



Characterization of nanoparticles in food and biological matrices by ICPMS based methods

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Environmental and Occupational
Health Aspects Related to

NANO- AND ULTRAFINE PARTICULATE MATTER

3-6 JUNE, 2019

HOTEL ALEXANDRA, LOEN, NORWAY





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Dear colleagues,

On behalf of the National Institute of Occupational Health in Norway, the Norwegian Institute of Public Health, the North-West Russian Public Health Research Centre, the Chemical Environmental Section of the Norwegian Chemical Society and the Organising Committee it is an honour and pleasure for me to welcome you to Loen at the EOHNANO 2019. The main aim of this conference is to provide a forum to encourage the exchange of ideas and knowledge about how ultrafine and nano-sized particulate matter may affect human health and the environment. The plenary programme features an exciting selection of high-level presentations of the latest developments in this scientific field. Since some of the world's leading authorities will be present, it is to be hoped that the conference will be a stimulating forum for communication across borders and between scientific disciplines.

We know that the natural beauty of the area will captivate you, but we also hope that the visit to Briksdalen with the outdoor farewell dinner may complement the scientific endeavours. We promise you an exciting and memorable conference and stay in Loen!

Balazs Berlinger
Chairman



ORGANISING AND SCIENTIFIC COMMITTEE

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PROGRAMME

MONDAY, JUNE 3

09:00	OPENING OF THE CONFERENCE
09:30	HEALTH EFFECTS OF NANO- AND ULTRAFINE PARTICLES (1) Chair: Karl-Christian Nordby
09:30	O-1 Status of epidemiological research on nanomaterial workers <i>Paul A. Schulte, Veruscka Leso, Mamadou Niang, Ivo Iavicoli</i>
10:15	O-2 Inhaled particles and the cardiovascular system Mark R. Miller
11:00	BREAK
11:30	O-3 Exposure assessment and health effects of different types of carbon nanotubes <i>Liliya Fatkhutdinova, Guzel Timerbulatova, Sergey Boychuk, Anna Antsiferova, Pavel Kashkarov, Shamil Zaripov, Timur Khaliullin, Ayrat Dimeev, Alexey Noskov, Albert Gilmutdinov, Yngvar Thomassen, Anna Shvedova</i>
12:15	O-4 The effect of nanomaterials on the surface tension of lung surfactant <i>Jorid B. Sørli, Emilie Da Silva</i>
12:35	O-5 Exposure to particles <250 nm in Norwegian smelters and endothelial function <i>Merete Drevvatne Bugge, Bente Ulvestad, Balazs Berlinger, Leo Stockfelt, Raymond Olsen, Dag G. Ellingsen</i>
12:55	LUNCH
14:00	HEALTH EFFECTS OF NANO- AND ULTRAFINE PARTICLES (2) Chair: Karl-Christian Nordby
14:00	O-6 Health impact of ultrafine particles on metabolic disease <i>Annette Peters</i>
14:45	SHORT BREAK
14:50	CHARACTERIZATION OF NANO- AND ULTRAFINE PARTICLES Chair: Balazs Berlinger
14:50	O-7 Characteristics of carbonaceous ultrafine particles – implications for toxicology and exposure assessment <i>Joakim Pagels</i>
15:35	BREAK



- 16:05 **O-8 Ultrafine particulate matter measurements and ultrafine particle characterization in occupational environment**
Balazs Berlinger, Torunn Kringlen Ervik, Yngvar Thomassen
- 16:50 **O-9 Chemical characterization of airborne nanoparticles in an industrial scenario**
Mar Viana, Apostolos Salmatonidis, Carla Ribalta, Eliseo Monfort, Sonia Fraga, Joao Paolo Teixeira, Flemming Cassee
- 17:10 **O-10 Toxic effects and characterization of gunshot fumes from different ammunitions for small arms**
Espen Mariussen, Lise Fjellsbø, Thomas Frømyr, Ida Vaa Johnsen, Øyvind Voie
- 17:30 POSTER SESSION**
- 19:00 DINNER**

TUESDAY, JUNE 4

- 09:00 EXPOSURE TO NANO- AND ULTRAFINE PARTICLES**
 Chair: **Yngvar Thomassen**
- 09:00 **O-11 Occupational exposure to nanoparticles – recognition, assessment and control at workplaces**
Anna-Kaisa Viitanen
- 09:45 **O-12 Environmental tobacco smoke exposure and the impact of a smoking ban in internet cafes, pubs, and billiard halls around a university in South Korea**
Kwonchul Ha
- 10:05 **O-13 Exposure assessment to airborne particles in an industrial workplace: The case study of pouring TiO₂ and filler materials for paint production**
Ana Sofia Fonseca, K. A. Jensen, A. Säämänen, T. Kanerva, A. -K. Viitanen, O. Aguerre-Chariol, C. Delpivo, A. J. Koivisto
- 10:25 **O-14 Daily time activities affecting black carbon (BC) exposure among elementary school children in South Korea**
Donguk Park, Hyeran Jeong
- 10:45 **O-15 Characterization of occupational exposures to fine and ultrafine particles in different workplaces in Quebec**
Maximilien Debia, Alan Fleck, Gilles L'Espérance, Caroline Couture
- 11:05 BREAK**



11:35 MEASUREMENT METHODOLOGIES (1)

Chair: **Yngvar Thomassen**

11:35 O-16 New instruments for direct sampling and real-time analysis of particulate organic matter in air

Armin Wisthaler, Markus Müller

12:20 O-17 Particulate matter measurements during in-situ burning of crude oil

Ingrid Gjesteland, Bjørg Eli Hollund, Jorunn Kirkeleit, Magne Bråtteit

12:40 O-18 Filtration of particles of traffic-origin attenuates their effects on blood pressure under controlled exposure conditions

Doug Brugge, Neelakshi Hudda, Misha Eliasziw, Ellin Reisner, Wig Zamore, John Durant

13:00 O-19 Certified laboratory analysis of carbon nanotube concentrations as elemental carbon

Patrick O'Shaughnessy, Craig Holder, Adrienne Stoltenberg, Ralph Altmaier

13:20 LUNCH

14:30 RISK ASSESSMENT AND MITIGATION OF EXPOSURE

Chair: **Håkan Wallin**

14:30 O-20 Characterization needs on nanomaterials for REACH registration in 2020 - current status of supporting characterization methods

Keld Alstrup Jensen

15:15 O-21 The contribution of personal monitors and samplers to occupational risk assessment of engineered nanomaterials

Luca Fontana, Ivo Iavicoli, Sergio Iavicoli

15:35 O-22 Safe production and use of nanomaterials in the ceramic industry: overview of results from the CERASAFE project

Mar Viana, Eliseo Monfort, Vicenta Sanfélix, Apostolos Salmatouidis and the CERASAFE consortium

15:55 BREAK

16:30 O-23 Occupational exposure modelling tools: requirements and lessons learnt

Ana Sofia Fonseca, A. J. Koivisto, C. Delpivo, R. A. Franken, A. Säämänen, T. Kanerva, A. - K. Viitanen, K. A. Jensen

16:50 O-24 Tools for risk assessment of nanomaterials at the workplace

Maike Visser, Daan Huizer, Susan Dekkers, Monique Groenewold

17:10 – 17:30 O-25 New indoor air quality standards; ISO16000-34: strategies for the measurement of airborne particles and ISO16000-37: measurement of PM2.5 mass concentration

Samuli Pylkkönen

19:00 DINNER



WEDNESDAY, JUNE 5

09:00 TOXICOLOGICAL MECHANISM

Chair: **Per Everhard Schwarze**

9:00 **O-26** Olfactory and central neurotoxicity of occupationally-relevant particulate aerosols and nanomaterials

Krishnan Sriram

9:45 **O-27** Particle cell interactions in the alveolar region of the lungs and their relevance for associated health effects

Tobias Stöger

10:30 **O-28** Evaluation of particle mass, number and volume specific surface area as dose metrics for internalization of nanoparticles

Fazel Abdolahpur Monikh, Daniel Arenas-Lago, Benjamin Fryer, Martina G. Vijver, Eugenia Valsami-Jones, Willie J. G. M. Peijnenburg

10:50 **BREAK**

11:20 **O-29** Resolving molecular events by super-resolution microscopy exemplified on interaction of TiO₂ nanotubes with cell layers in live

Hana Majaron, Aleksandar Sebastijanović, Boštjan Kokot, Patrycja Zawilska, Tilen Koklič, Janez Štrancar

12:05 **O-30** Nanomaterials accelerate systemic autoimmune disease in lupus-prone New Zealand mixed mice

Andrij Holian

12:25 **O-31** MWCNT inhalation toxicity – influence of aerosol characteristics

Chang Guo, Alison Buckley, Matthew Wright, Sarah Robertson, Alan Hodgson, Martin O. Leonard, Timothy W. Gant, Rachel Smith

12:45 **O-32** Important inferences from animal experiments assessing adverse health effects of metal oxide nanoparticles

M. P. Sutunkova, B. A. Katsnelson, L. I. Privalova, I. A. Minigalieva, V. B. Gurvich, S. N. Soloveva, S. V. Klinova, O. G. Makeyev, I. E. Valamina, R. R. Sakhautdinova, T. V. Bushueva, V. Y. Shur, I. V. Zubarev, E. V. Shishkina

13:05 **LUNCH**

14:30 **MEASUREMENT METHODOLOGIES (2)**

Chair: **Yngvar Thomassen**

14:30 – 15:15 **O-33** Characterization of nanoparticles in food and biological matrices by ICP-MS based methods

Katrin Loeschner

16:00 **EXCURSION TO BRIKSDAL GLACIER AND CONFERENCE OUTDOOR DINNER**

THURSDAY, JUNE 6

9:00 **PANEL DISCUSSION** with the invited speakers

Moderators: **Karl-Christian Nordby, Per Everhard Schwarze and Håkan Wallin**

10:15 **BREAK**

10:45 **PANEL DISCUSSION (cont.) AND CLOSING REMARKS**

12:00 **LUNCH**



POSTERS

P-1 Toxicity evaluation of airborne ceramic thermal spraying-generated nanoparticles in a 3D human bronchial epithelial model

Maria João Bessa, Fátima Brandão, Apostolos Salmatonidis, Adriana Vulpoi, Mar Viana, Flemming Cassee, Sónia Fraga, João Paulo Teixeira

P-2 Indoor and outdoor particle number concentration in the Sapienza University campus of Rome

Raffaella Gaddi, Giorgio Cattani, Mariacarmela Cusano, Alessandro Di Menno di Bucchianico, Alessandra Gaeta, Gianluca Leone, Fabio Boccuni, Riccardo Ferrante, Armando Pelliccioni

P-3 Road-motor traffic complex as a source of polyaromatic hydrocarbons contained in fine dust particles

S.A. Gorbanev, O.I. Kopytenkova, A.V. Levanchuk

P-4 Formation of ultrafine particles in a classroom under different ventilation conditions

Claudia Hak, Matthias Vogt, Franck Dauge, Øystein Fjellheim, Sverre Holøs, Aileen Yang, Tomas Mikoviny, Armin Wisthaler

P-5 Non-invasive sensitive method for oxidative stress quantification in exhaled air

M. Hemmendinger, N. Sambiagio, S. Goekce, N. Concha-lozano, I. Guseva Canu, G. Suarez, J.J. Sauvain

P-6 Study findings of chemical composition of fine dust particles in the area of road-motor traffic complex exposure

Levanchuk Aleksandr, Kopytenkova Ol'ga, Gennadii Eremin

P-7 Characterisation of air contaminants emitted during laser cutting of carbon fiber reinforced composite materials

Alexey Noskov, Albert Gilmutdinov, Yngvar Thomassen

P-8 Processing of nanocomposites and grouping of exposure

Bianca Oeffling, Carsten Möhlmann, Christian Schumacher, Denis Goldnik, Birgit Funk, Andreas Klingler, Alla Zak

P-9 Does exposure to inflammatory particles modify the pattern of anions in exhaled breath condensate?

Jean-Jacques Sauvain, Guillaume Suarez, Jean-Louis Edmé, Pascal Wild, Olivia Bezerra, Keller Silveira, Lénio Amaral, Anna Paula Carneiro, Nathalie Chérot-Kornobis, Annie Sobaszek, Sébastien Hulo

P-10 Exposure to unintentional nanoparticles during thermal spraying

Apostolos Salmatonidis, Carla Ribalta, Spyros Bezantakos, George Biskos, Vicenta Sanfelix, Eliseo Monfort, Mar Viana

P-11 Analysis of titanium dioxide nanoparticles in food by single particle ICP-MS/MS

Janja Vidmar, Sara López-Sanz, Laurent Devuille, Johanna Noireaux, Manuel Correia, Katrin Loeschner

ABSTRACTS

O-1 STATUS OF EPIDEMIOLOGICAL RESEARCH ON NANOMATERIAL WORKERS

Paul A Schulte¹, Veruscka Leso², Mamadou Niang³, Ivo Iavicoli²

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International concern about the potential health effects of engineered nanomaterials (ENM) has led to the conduct of extensive research on the health effects of these nanomaterials since the early 2000's. Most of that research involved studies of laboratory animals. For workers, there have been fewer studies; the majority being published in the last 5 years. Now is a good time to review the published epidemiologic and human studies and discuss what is known about health effects from ENM in workers. The published scientific literature for the period 2003-2016, was systematically reviewed to identify human studies and epidemiologic investigation of ENM workers exposed to the ten ENMs most found in commerce. The review was conducted using major scientific data bases including Web of Science, Scopus, and PubMed. The primary selection criterion for epidemiological studies was that they included the use of a non-exposed or comparison group. Twenty-seven studies were identified. Most of the epidemiological investigations were cross-sectional. The review found limited evidence of adverse effects in workers exposed to the most commonly used ENM. However, some biological alterations are suggestive for possible adverse impacts. The primary targets of some ENM exposures were the respiratory and cardiovascular systems. Although the mechanisms for ENM-induced health effects are not fully known, the persistent inflammatory and related oxidative stress reactions induced by ENM may determine alterations in lung functionality, as reported in workers exposed to carbon black or to TiO₂-ENM. In line with these results, inflammatory changes in biomarker levels compared with controls were also observed; however, limited exposure data and the relatively short period since the first exposure may have influenced the incidence of adverse effects found in epidemiological studies. Various findings are highly suggestive of the potential for cardiovascular dysfunction resulting from a systemic inflammatory status following pulmonary exposure. The study illustrated the need for longitudinal epidemiologic investigations with clear exposure characterizations for various ENM to discover potential adverse health effects and identify possible indicators of early biological alterations. In this state of uncertainty, precautionary controls for each ENM are warranted while further study of potential health effects continues.

O-2 INHALED PARTICLES AND THE CARDIOVASCULAR SYSTEM

Mark R. Miller

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Over the last few decades, it has become apparent that particles in air pollution have many detrimental effects in different organ systems. The cardiovascular effects of inhaled particles are especially important, given the high proportion of deaths attributed to air pollution that arise from cardiovascular causes. Vehicle exhaust is an especially rich source of nanoparticles in urban air pollution.

This presentation shall provide an overview of the Edinburgh Air Pollution Programme, studying the cardiovascular effects of diesel exhaust particles using controlled exposures in human subjects and mechanistic work in preclinical models. The presentation will discuss the potential for particle translocation (the movement of particles from the lung into the circulation) to account for the widespread cardiovascular actions in inhaled nanoparticles.

Diesel exhaust caused a striking array of detrimental actions on different facets of the cardiovascular system. Acute exposure in man promoted blood clotting, impaired vascular function, and made the heart more susceptible for ischaemic stress. *In vivo* rodent models demonstrated that repeated exposure to diesel exhaust particulate exacerbated the chronic vascular disease atherosclerosis and identified a number of different biological pathways including inflammation, oxidative stress and changes to the autonomic nervous system.

While there is strong evidence for particle translocation in animal models, the evidence for this pathway in man is limited, and the cardiovascular implications have received little attention. We have performed a series of translational investigations using gold nanoparticles as a model to study particle translocation. Following inhalation of gold nanoparticles in healthy volunteers, we were able to demonstrate the presence of gold in the blood and urine within hours of exposure, which then persisted for a number of months. The translocation of gold nanoparticles was size-dependent, with smaller particles more readily entering the blood. Using a mouse model of atherosclerosis, we demonstrated that translocated nanoparticles preferentially accumulated at areas of vascular disease. Finally, we demonstrated that inhaled particles reached areas of atherosclerosis in patients with a history of stroke.

Our research provides insight into the means by which inhaled ultrafine particles can promote cardiovascular morbidity and mortality. Particle translocation represents a convincing underpinning mechanism for the multiple detrimental effects of inhaled nanoparticles. As well as having important implications for the nanomaterial industry, these findings emphasise the need to reduce emissions from traffic as an important means to reduce the health effects of air pollution.

O-3 EXPOSURE ASSESSMENT AND HEALTH EFFECTS OF DIFFERENT TYPES OF CARBON NANOTUBES

Liliya Fatkhutdinova¹, Guzel Timerbulatova¹, Sergey Boychuk¹, Anna Antsiferova², Pavel Kashkarov², Shamil Zaripov³, Timur Khaliullin⁴, Ayrat Dimeev³, Alexey Noskov⁵, Albert Gilmutdinov⁵, Yngvar Thomassen⁶, Anna Shvedova⁴

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Carbon nanotubes (CNT) production is steadily increasing due to growing demands in construction, engineering, energy-producing, space exploration and biomedicine. At the same time, experts and consumers raise questions worldwide on the safety of CNTs. These carbonaceous nanostructures may be released into the workplace air during manufacturing and handling, composite materials wear and tear, or as by-products in a variety of technological processes. Ambient air contamination may occur in metropolitan areas, as a part of the motor fuel combustion and forest fires.

We are still far from the consensus on the most appropriate method of assessing occupational/incidental exposure to CNTs. Measuring mass concentrations of elemental carbon is currently one of the preferred methods, but there are issues associated with (1) the use of various modifications of the method, (2) difficulties differentiating actual carbon nanotubes from other carbonaceous particles that may be present in the environment. Other approaches are proposed, such as using the surface-enhanced Raman spectroscopy for determination of airborne carbon nanotube content in workplaces. The study of by-products and environmental levels evaluation are even more challenging. Here we present several examples of CNT exposure assessment studies, including verification of the possible presence of similar structures at various industrial sites.

In addition to the exposure assessment, a deeper understanding of the CNT biokinetics and bioaccumulation is important for finding target organs and verifying dose-response relationships. Despite more than a decade of research, transportation and biodistribution of carbon nanotubes in mammalian organisms remain unclear. To quantitatively determine the content of carbon nanotubes in the biological tissues of laboratory mammals, as well as in human samples, we are proposing the method of instrumental neutron activation analysis based on radioactive labelling of well-activated catalyst residues in the carbon nanotubes.

We would also discuss the methodological challenges that we have encountered during different types of toxicity studies (stable CNT dispersions preparation, *in vitro* models, dose selection, epidemiological studies).

The study was partly supported by the Russian Foundation for Basic Research (project No 18-32-20197).

O-4 THE EFFECT OF NANOMATERIALS ON THE SURFACE TENSION OF LUNG SURFACTANT

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The presence of lung surfactant (LS), which covers the most of the lung epithelium including the respiratory parts of the lungs namely the alveoli and the respiratory bronchioles, is vital for normal respiration. LS regulates the surface tension of the lung lining fluid at the interface between air and liquid and the underlying tissue, which keeps the alveoli from collapsing during respiration. A disruption of this important function of the LS can lead to acute inhalation toxicity, with symptoms such as coughing, tightness in the chest and problems with breathing. Ultimately, this can lead to long-term problems with the lungs, such as fibrosis, although these effects are harder to characterize. How nanomaterials (NMs) affect LS function in a cell-free in vitro setup was explored by testing 17 NMs that cover a broad range of materials, several with the same core material, but with different surface modifications. The LOAEL (lowest observed adverse effect level) was determined for most materials, by mixing LS with increasing doses of NMs until a statistically significant effect was seen on minimum surface tension. The LOAEL was used to rank the materials from having the strongest effect on lung surfactant function, to those that did not affect the function. For surface modified materials, the modification had a great effect on the LOAEL. The tested materials include carbon materials in different forms and metal oxides.

O-5 ENDOTHELIAL FUNCTION AMONG WORKERS EXPOSED TO PARTICLES <250 NM IN NORWEGIAN SMELTERS

Merete Drevvatne Bugge¹, Bente Ulvestad¹, Balazs Berlinger¹, Leo Stockfelt², Raymond Olsen¹, Dag G. Ellingsen¹

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Objectives

Exposure to fine particles (PM_{2.5}) in urban air has been associated with increased risk of hospitalization and death from cardiovascular diseases. Few studies of cardiovascular effects from fine particles have been performed in a working environment. The endothelium is an important transmitter of signals in the cardiovascular system, and dysfunction of the endothelium is an early marker of increased risk of cardiovascular disease. The aim of this study was to examine the association between exposure to respirable and ultrafine particle size fractions in Norwegian smelter plants and endothelial function.

Methods

We examined 59 workers in three Norwegian metal smelters after a working day in the furnace hall, and after at least two consecutive days off work. On the working day, exposure to different particle size fractions was assessed with personal sampling using respirable cyclones and five-stage Sioutas cascade impactors. Baseline pulse amplitude and endothelial function were measured with Endo-PAT2000 (Itamar Medical Ltd.). Associations between the mass of different particle size fractions and outcomes were assessed using fixed effects linear regression.

Results

Endothelial function measured by reactive hyperemia index was reduced on a working day relative to a day off. No associations, however, were observed with increasing exposure to respirable particles or particles <250 nm. Baseline pulse amplitude was increased on a working day relative to a day off, and significant exposure-response associations were observed with particle exposure among the older half of the participants (>34 years).

Conclusion

Exposure to respirable and ultrafine particles in Norwegian smelters was associated with increased baseline pulse amplitude among the older workers.

O-6 HEALTH IMPACT OF ULTRAFINE PARTICLES ON METABOLIC DISEASE

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O-7 CHARACTERISTICS OF CARBONACEOUS ULTRAFINE PARTICLES -IMPLICATIONS FOR TOXICOLOGY AND EXPOSURE ASSESSMENT

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Exposure to airborne particles (PM_{2.5}) is associated with a number of adverse health effects in the population. There is increasing evidence that ultrafine particles (particles < 100 nm) has additional health effects (Downward et al. 2018). Diesel exhaust (DE) is a major source to UFPs. DE is classified as carcinogenic (IARC 2012) and EU recently agreed on future OEL's in the working environment based on Elemental Carbon (EC). Nanotoxicological studies have established particle surface area as a major determinant of toxicological effects, meaning that the toxicity per mass unit increases with decreasing particle size. The respiratory tract deposition in the alveolar region of the lung is highest for the 5-50 nm particles. Engineered solid carbon black particles have been shown to be genotoxic. An important group of genotoxic compounds in combustion particles are polycyclic aromatic hydrocarbons (PAH).

Diesel exhaust commonly contains two main types of particles: 1. Insoluble EC dominated 50-100 nm aggregates of smaller primary particles, formed during the combustion process in the engine (Malmborg et al. 2017). 2. Smaller nucleation mode particles (5-20 nm) that predominantly consist of soluble material (e.g. lubrication oils) and form mainly after the tail-pipe. Commonly, the larger sized mode dominates the mass concentration and the smaller sized mode dominates the number concentration. Modern after treatment systems such as diesel particulate filters (DPF) reduce emissions and change the particle properties.

Biomass combustion emissions can be approximated to consist of three major particle types (Bølling et al. 2009): 1. Insoluble EC dominated particles with aggregate sizes 100-150 nm from wood stoves and other well-insulated systems, 2. Organic aerosol (e.g. anhydrous sugars and phenols) dominated particles (50-200 nm) from combustion with moist fuel, poorly insulated systems and from forest fires. These conditions lead to the highest PM_{2.5} emissions, 3. Modern technologies such as wood pellet systems can have low carbonaceous emissions and low PM_{2.5} emissions. Emitted particles (50-100 nm) are then dominated by water-soluble potassium chlorides and sulphates and are enriched with Zinc. Combustion conditions leading to strongly elevated PAH emissions have been established (Nielsen et al. 2017)

When combustion emissions age in the atmosphere (hours to days) gas-phase emissions are oxidized and additional particle mass is formed. Examples are NO_x in diesel exhaust that forms particulate nitrate and light aromatic compounds in gasoline exhaust that form secondary organic aerosol (Nordin et al. 2013). Secondary PM often dominates over primary PM and is a major source of long-range transport PM_{2.5}. A majority of the secondary components are water-soluble.

In the talk, the implications of those findings for toxicology (specific surface area, surface properties, solubility) and exposure assessment (measurement techniques) will be discussed.

Acknowledgements: Swedish Research Councils FORMAS and VR, AFA Insurance

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O-8 ULTRAFINE PARTICULATE MATTER MEASUREMENTS AND ULTRAFINE PARTICLE CHARACTERIZATION IN OCCUPATIONAL ENVIRONMENT

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Exposure to airborne particulate matter (PM) is a major risk factor for development of respiratory and cardiovascular diseases. Epidemiological and experimental studies have shown that particle size and physical and chemical properties of the PM are of importance, with possible increased toxicity of the smallest particles. Many industrial workers are exposed to agglomerates/aggregates composed of primary ultrafine particles ($<0.1\ \mu\text{m}$). These primary particles are unintentionally formed, e.g. in thermal processes used in the production of metals/alloys, by welding and by mechanical grinding of clinker in cement factories.

The aim of the present study is to characterize the PM in workplace air during various industrial processes with regard to particle size distribution, morphology, chemical composition and solubility/bioavailability. PM formed in recycling and casting of precious metals, production of cement, aluminum, silicon, silicon carbide, ferroalloys (silico- and ferromanganese, ferrosilicon) and nano zinc metallic powder; in different welding techniques and other hot processes such as plasma and thermal cutting and air carbon arc applications are under characterization.

The applied sampling and analytical methods include the use of personal and stationary cascade impactors for subsequent characterization of the particle fractions by gravimetric analysis, electron microscopy (SEM, ESEM, TEM) and atomic spectrometry (ICP-OES, ICP-MS).

In this presentation the characteristics of PM present in workroom air in different industries with respect to particle size (mass and number) distribution, morphology, chemical composition and solubility/bioavailability will be discussed.

O-9 CHEMICAL CHARACTERIZATION OF AIRBORNE NANOPARTICLES IN AN INDUSTRIAL SCENARIO

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Atmospheric plasma spraying is applied at industrial scale to produce high-resistance coatings of metallic surfaces. Due to the high energies applied, it is a known source of nanoparticles (NP) which are released to workplace air and impact worker exposure in industrial facilities. NP emissions in this kind of scenario are characterized in terms of particle number concentrations, mean diameter and size distribution, but data are lacking regarding the chemical properties and toxicity of these particles.

In this framework, this work aimed to characterize the chemical composition of unintentionally-emitted fine and ultrafine aerosols emitted during plasma spraying in a real-world setting, with the ultimate goal to support ongoing toxicity assessments of the same aerosols. Two different scenarios (Booths #1 and #3) were assessed. Particles were sampled onto Teflon filters and in suspension using an aerosol concentration enrichment system (VACES; Kim et al., 2001), and analyzed by means of ICP-MS, ICP-OES and XRF. Results evidenced a major enrichment in potentially health hazardous metals (Cr, Ni, W) sourcing directly from the feedstock in both scenarios, as well as in major elements (Al, Ca, Fe) with different possible source origins (including outdoor infiltration). As, Ni and Cr showed the highest enrichments in the ultrafine fraction in Booth#1, while in Booth #3 metals were mainly present in the fine aerosol fraction probably due to the different type of spraying used. Aerosol chemical properties were correlated with bulk material composition, to understand the relationship between bulk composition and NP emissions in the three size fractions analysed. Toxicity assessments will provide further quantitative insights into the health hazards of this kind of exposure.

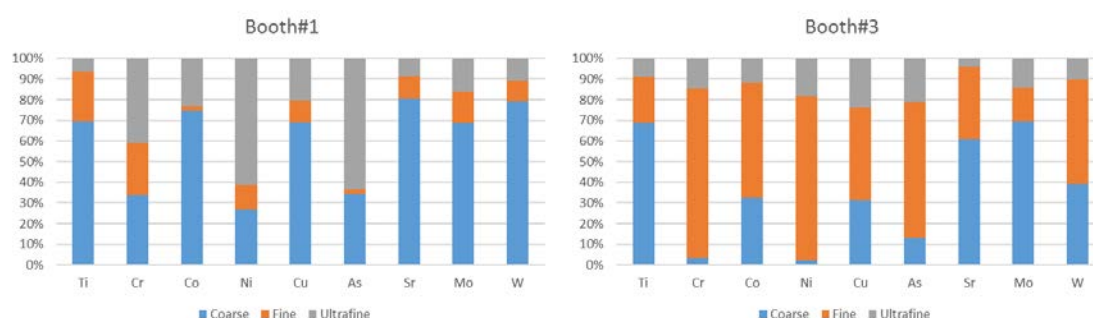


Figure. Relative contribution of metals to the different aerosol fractions in both scenarios.

O-10 TOXIC EFFECTS AND CHARACTERIZATION OF GUNSHOT FUMES FROM DIFFERENT AMMUNITIONS FOR SMALL ARMS

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Gunshot fumes from firing small arms contain a complex mixture of gases, particles and aerosols. Exposure to gunshot fumes may induce health effects such as fever, coughing, inflammation, and reduced lung capacity. These are symptoms resembling metal fume fever. Concerns are raised if gunshot fumes may induce more long lasting effects, such as prolonged reduced lung capacity and even cancer. The toxic effect of the gunshot fumes is related to the chemical composition of the smoke and the formation of particles, of which the particle size distribution of the smoke is of particular importance. In this study, the particle size distribution of gunshot fumes from three different ammunition types were characterized by an electrical low-pressure impactor and a Fast Mobility Particle Sizer (FMPS). Each filter from the impactor was subjected to chemical analysis of Cu, Zn and Pb. The genotoxic potential of the gunshot fumes on human lung cells exposed in an air liquid interface (ALI) was, in addition investigated. The genotoxicity was elucidated by the comet assay, which is a widely used method to detect DNA breaks in vitro as well as in vivo. A modified versions of the comet assay was also used to detect oxidized purines, which is an indicator of oxidative stress induced DNA lesions. The cytotoxic effect of the smoke were tested to compare the general toxicity between the different ammunition types.

The particle characterization of the gunshot fumes revealed that, irrespectively of ammunition type, the highest mass fraction of particles had a size between 1-3 μm . The unleaded ammunition NM229 also generated a large mass fraction of approximately 250 nm. The emission of Cu, Zn and Pb showed approximately the same pattern as the particle mass distribution with a peak around 2 μm . The highest number of particles, however, generated immediately after by the firing of gun shots were in the size range of approximately 35 nm, which within 60 seconds agglomerated to a size of approximately 60 nm. Particles less than 100 nm readily penetrate into the alveolar regions of the lung. Lung epithelial cells were exposed in ALI to the fumes. Fumes from the unleaded ammunitions, NM255 and NM229, induced cytotoxic effects of which the emission from NM229 apparently was the most toxic. The higher cytotoxicity of NM229 could be attributed to a higher amount of particles deposited on the cells. Only the emission from NM229 induced a dose-related increase in DNA-damage. Significant effects were only achieved at the highest exposure level, which eventually would lead to reduced cell viability after 24 hours of approximately 40%.

The results indicates that acute exposure to high concentrations of gunshot fumes can induce cytotoxic distress and even genotoxic effects. The effect probably relates to the amount of emitted particles of which their size may be of importance. A complex mixture of chemical substances in addition to the particles may increase the toxicity of the fumes, and one should merit measures to reduce exposure during training.

O-11 OCCUPATIONAL EXPOSURE TO NANOPARTICLES – RECOGNITION, ASSESSMENT AND CONTROL AT WORKPLACES

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Occupational exposure to nanoparticles may be divided to the exposure to incidental and to engineered nanoparticles. Incidental nanoparticles are being formed as a side product of different processes and work phases, and relatively high concentrations have been measured in various environments from offices to industry. Especially processes with high temperature and energy are shown to produce high amounts of incidental nanoparticles which workers' may be exposed to.

On the other hand, nanoparticles are also being produced on purpose for different industrial applications, i.e. engineered nanoparticles. Occupational exposure to engineered nanoparticles may occur when engineered nanoparticles or nanoenabled products are being synthesized or handled during different work tasks and processes. Especially handling of powder form nanomaterial as well as cleaning and maintenance tasks are shown to pose a risk for occupational exposure.

The nanoparticle concentrations from the workplace air may be determined by using different on-line and off-line methods. The mass of nanoparticles is extremely low and hence, the methods based on the measurement of the particle number or surface area (SA) concentrations are typically being used for exposure assessment. There are no occupational exposure limit values given for the number or SA concentrations nor is the toxicity of various nanoparticles well known which sets a challenge for the overall risk assessment.

Since nanoparticles may be released during different work tasks and from different processes, the whole production line should be involved when assessing the workers' exposure to nanoparticles. There are number of different means and control methods available for managing the risk for nanoparticle exposure. These methods including Safe-by-Design approach in risk management are being discussed along with the levels of nanoparticle concentrations at different work environments, different sources and exposure assessment practices.

The results presented are partly achieved from the European Union's Horizon 2020 funded projects NECOMADA (No 720897) and EC4SafeNano (No 723623).

O-12 ETS EXPOSURE AND THE IMPACT OF A SMOKING BAN IN INTERNET CAFES, PUBS, AND BILLIARD HALLS AROUND A UNIVERSITY IN SOUTH KOREA

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The objectives of this study were to determine indoor levels of environmental tobacco smoke (ETS) and assess the implementation rate of a smoking ban in hospitality venues surrounding a university campus by measuring particulate matter smaller than 2.5 μm ($\text{PM}_{2.5}$) as an indicator of ETS. We identified the smoking state in business establishments and measured the indoor $\text{PM}_{2.5}$ concentrations at 20 internet cafes, 38 pubs, and 20 billiard halls using a Sidepak AM510 direct-reading portable real time monitor from October 2014 to December 2015. Smoking was observed in 65% of the internet cafes and 85% of the billiard halls in 2015. The rate in pubs, which were subject to a legal smoking ban, was reduced to 10% in 2015 from 33.3% in 2014. The average $\text{PM}_{2.5}$ concentrations in 2015 were 98.6 $\mu\text{g}/\text{m}^3$, 29.6 $\mu\text{g}/\text{m}^3$, and 135.4 $\mu\text{g}/\text{m}^3$ in internet cafes, pubs, and billiard halls, respectively. $\text{PM}_{2.5}$ concentrations in internet cafes and billiard halls were 2 to 2.7 times higher than the 24-hour exposure standard (50 $\mu\text{g}/\text{m}^3$) for outdoor $\text{PM}_{2.5}$ set by the Korean Ministry of Environment. Although a smoking ban had been imposed on internet cafes and pubs, smoking was still taking place in those locations. More stringent enforcement is required for the success of legal measures to protect patrons' and workers' health from secondhand smoke exposure. A ban on smoking in billiard halls should be introduced as soon as is feasible.

Reference

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O-13 EXPOSURE ASSESSMENT TO AIRBORNE PARTICLES IN AN INDUSTRIAL WORKPLACE: THE CASE STUDY OF POURING TiO₂ AND FILLER MATERIALS FOR PAINT PRODUCTION

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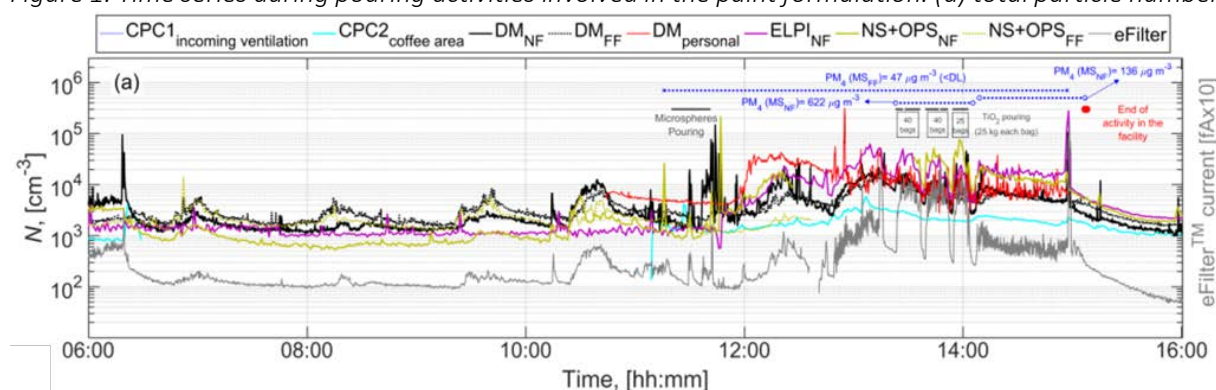
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Exposure to micro- and nanoscaled particles has been widely linked to adverse health effects including pulmonary, cardiovascular and nervous system diseases. Therefore, occupational exposure assessment studies should be conducted to investigate whether workers health is at risk. Industrial case studies are also needed to generate new occupational exposure and environmental release data for demonstration of modelling tools. In this study, a field campaign was conducted at a paint manufacturer, during production of paint batches.

The measurement plan included real time particle monitoring, collection of particles in filter samplers and TEM samples from near field, far field, breathing zone, and outdoors in the size range of 5 nm to 30 µm by using a variety of instruments.

Relevant personal exposure levels in terms of mass, number and lung deposited surface area concentrations were measured during pouring activities (Figure 1). Used TiO₂ and filler materials were always detected by TEM analysis in the samples collected indoors and in the stack emissions. In particular, the dust collected during kaolin and talc pouring activities were dominated by micrometric platelets. Considering the 8h-time weighted average (8h-TWA), none of the existing recommended exposure limits were exceeded. However due to concentrations > 1 mg/m³, it is recommended to increase the efficacy of the exposure management measures. In addition, work practices and procedures should be in place to prevent exposure and to minimize particle generation. Suitability of the gathered exposure measurement data for risk assessment with different models and tools will be discussed.

Figure 1. Time series during pouring activities involved in the paint formulation. (a) total particle number



concentration.

Acknowledgments

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O-14 DAILY TIME ACTIVITIES AFFECTING BLACK CARBON (BC) EXPOSURE AMONG ELEMENTARY SCHOOL CHILDREN IN SOUTH KOREA

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A daily black carbon (BC) exposure assessment of 40 children aged 10-12 was conducted in the Seoul from August 2015 to January 2016. Each participant carried a micro-aethalometer to measure BC concentrations for 24 hours while their whereabouts and microenvironments (MEs) were recorded via a time-activity diary (TAD) and follow-up interviews. Analysis of variance (ANOVA) was employed to compare average BC levels by potential risk factors including demographic, temporal, residential, and indoor/outdoor/transportation activity variables. Potential inhaled dose was estimated by multiplying the airborne BC concentrations ($\mu\text{g}/\text{m}^3$) we monitored for the time the children spent in a particular ME by the inhalation rate (IR, m^3/h) for the time-activity performed. The contribution of activities and MEs to overall daily exposure to and potential dose of BC was quantified. The children's average daily exposure was $1.93 \mu\text{g}/\text{m}^3$, with a range of $0.2\text{--}85.43 \mu\text{g}/\text{m}^3$ (mean daily individual exposure ranges from 0.54 to $4.80 \mu\text{g}/\text{m}^3$). Even children attending the same elementary school reported BC exposures which differed by approximately 40%, primarily because of individually distinct time-activity patterns and the MEs with which each child interacted. On weekends (Saturdays and Sundays) ($1.86 \pm 2.50 \mu\text{g}/\text{m}^3$) and holidays (Saturdays, Sundays, and vacation) ($1.71 \pm 2.48 \mu\text{g}/\text{m}^3$), children were subject to reduced exposures to BC, likely due to decreased surrounding traffic volumes and different time-activity patterns on weekend days compared to on weekdays (from Mondays to Fridays) ($1.95 \pm 2.44 \mu\text{g}/\text{m}^3$) or school days (weekdays during the school semesters) ($2.05 \pm 2.43 \mu\text{g}/\text{m}^3$). The largest contribution to BC exposure and potential dose (51.9% and 41.7% respectively) occurred in the home thanks to the large amount of time spent there. Transportation was where children received the most intense exposure to (14.8%) and potential dose (20.2%) of BC, while it accounted for 7.6% of daily time. School on weekdays during the semester was responsible for 20.3% of exposure and 22.5% of potential dose. Commuting in diesel vehicles (often to private academies) or in the subway, cooking, and environmental tobacco smoke were all found to elevate BC exposure. Likewise, proximity to traffic sources and parental indoor smoking contributed to the enhancement of residential BC concentrations. Our findings suggested a need to emplace proactive measures including diesel fleet regulation and smoking cessation campaigns to protect children from high levels of BC exposure. This study may be used to prioritize targets for minimizing children's exposure to BC and to indicate outcomes of BC control strategies.

Reference

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O- 15 CHARACTERIZATION OF OCCUPATIONAL EXPOSURES TO FINE AND ULTRAFINE PARTICLES IN DIFFERENT WORKPLACES IN QUEBEC

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A similar intervention plan consisting of a combination of ambient direct real-time measurements and integrated measurements was performed in different workplaces (Québec, Canada) to characterize occupational exposures to fine and ultrafine particles (FP and UFP). Three of these workplaces were environments with the presence of equipment operating with diesel engines: an underground mine (W1), underground subway tunnels with the presence of diesel locomotives (W2) and a truck repair workshop (W3). The two other workplaces were a foundry (W4) and a machine shop that includes activities such as welding, grinding and cutting (W5).

Direct reading instruments (DRI)s were installed in two portable suitcases [a condensation particle counter (CPC Ptrak 8525, TSI Inc. for particle number concentration (PNC)), an optical particle counter (OPC) (DustTrak DRX, TSI Inc. for PM₁, PM_{resp}, PM₁₀) and a light-transmission monitor for elemental carbon submicron fraction (for EC₁; Airtec, FLIR Inc.)]. In addition, a set of different filters and size selectors for integrated measurements and characterization of dust mass concentration in different size fractions were used. Integrated measurements in W1, W2 and W3 assessed the respirable and submicron fractions of elemental carbon (EC_R and EC₁) and total carbon (TC_R and TC₁), while mass measurements (37-mm cassette and SIOUTAS impactor; SKC Inc.) followed by metal analysis (ICP-MS) were performed in W4 and W5. Finally, aerosol samples for subsequent analysis of morphology and chemical characterization by electron microscopy coupled with energy-dispersive X-ray spectroscopy were collected with the Mini Particle Sampler (MPS; ECOMESURE Inc). Measurements were performed for 6 days between 4 and 6 hours at each location, in a total of 12 measurements per workplace.

Daily geometric mean particle number concentrations ranged from 12,900 p/cm³ to 228,600 p/cm³. The three workplaces with the highest particle number concentrations were the underground mine (W1), the foundry (W4) and the machining shop (W5). Daily geometric mean mass concentrations (PM₁) ranged from 0.01 to 1.53 mg/m³ with the highest concentrations in W1. Significant seasonal variations (p<0.01) were observed for W3 and W4. Strong correlations were found between filter-based concentrations and DRIs' daily mean concentrations: EC₁-filter and EC₁-Airtec (r=0.97; p<0.001); EC₁-filter and PM_{resp}-DRX (r=0.94; p<0.001); EC₁-filter and PNC-CPC (r=0.86; p<0.001); metal respirable mass concentrations-filter and PM_{resp}-DRX (r=0.57; p<0.01) and between metal respirable mass concentrations-filter and PNC-CPC (r=0.47; p<0.05). Airborne particles in W4 were agglomerated chains of dozens of particles and mass median aerodynamic diameters (MMADs) of metals were > 1 µm. Particles generated in W5 during welding were large agglomerated chains of hundreds of particles with MMADs of metals < 1 µm. Particles generated in W5 during grinding and machining were coarser particles and corresponding MMADs of metals reached 2.4 µm. Airborne particles collected in W1, W2 and W3 were large chains of hundreds of carbon particles and large majority of particles had an aerodynamic diameter in the submicron fraction (<1 µm).

The strategy employed in this study made it possible to precisely characterize FP and UFP emitted in five workplaces in terms of particle number concentration, mass concentration, morphology and chemical composition.

O-16 NEW INSTRUMENTS FOR DIRECT SAMPLING AND REAL-TIME ANALYSIS OF PARTICULATE ORGANIC MATTER IN AIR

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Particle emission and exposure measurements often require a time resolution on the order of seconds. Commercial instruments are nowadays routinely used for monitoring particle mass and number concentrations as well as particle size distributions in real time. Fast measurements of the chemical composition and especially of the organic composition of submicrometer particles do, however, remain an analytical challenge. For two decades, the atmospheric science community has almost exclusively relied on the use of aerosol mass spectrometry (AMS) for chemically characterizing aerosol particles in real time. In AMS, organic molecules do, however, extensively fragment when analyte particles are vaporized at 600 °C and the emanating gases are ionized by 70 eV electrons. This has limited the applicability of AMS to organic particle analysis and driven the development of new online and real-time mass spectrometry techniques based on low-temperature analyte vaporization or extraction and soft chemical ionization. In my talk, I will focus on the two methods that have recently reached commercial maturity, extractive electrospray ionization time-of-flight mass spectrometry (EESI-ToF-MS) [1] and proton-transfer-reaction time-of-flight mass spectrometry (PTR-ToF-MS) [2]. The latter is a well-established technique in organic trace gas analysis and the development of the CHARON (*“Chemical Analysis of Aerosol Online”*) particle inlet in my research group has enabled it to be used for particle analysis [3]. I will present and compare the EESI-ToF-MS and CHARON PTR-ToF-MS techniques and show a variety of application examples. These include the characterization of primary organic particle emissions from a ship diesel engine and a wildfire, source apportionment of urban particulate organic matter and the detection of carcinogenic nitrosamines and potentially carcinogenic nitramines in particles generated by the photo-oxidation of industrially emitted amines. The advent of new particle measurement technologies for real-time organic monitoring is anticipated to open new avenues in environmental and occupational exposure research.

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O-17 PARTICULATE MATTER MEASUREMENTS DURING IN-SITU BURNING OF CRUDE OIL

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Introduction: Oil response personnel could be exposed to particulate matter (PM) during in-situ burning of crude oil at sea, which is being implemented as an oil spill response method. The aim was to investigate the concentration of different particle fractions at sea level during this process.

Methods: One North Sea crude oil (Oseberg) and one ultra-low sulphur fuel oil (ULSFO) were tested. Direct-reading instruments were used to measure PM_{<1}, PM₁, PM_{2.5}, PM₄ and PM₁₀ on two open boats located downwind during the burn. One boat (A) transected back and forth under the smoke close (200 m) to the oil slick, and the other boat (B) followed the smoke from the oil slick and downwind (0-3 km). Background measurements were performed on a large vessel located upwind.

Results: The crude oils produced nearly the same air concentration of the respective particle fractions above PM₁. The concentration was highest on boat A, with average PM_{2.5} concentrations of 0.065 mg/m³ (Oseberg) and 0.059 mg/m³ (ULSFO). Maximum concentration just below the smoke was 1.2 mg/m³. The average PM_{2.5} concentration was 0.006 mg/m³ (both burns) on boat B and 0.018 mg/m³ (Oseberg) and 0.029 mg/m³ (ULSFO) on the large vessel.

Discussion: Particle levels varied between the three boats and were highest just below the smoke, 200 m downwind of the oil slick. Still, significant amounts of particles were detected both upwind and far downwind (3 km). The results indicate that a large fraction of the smoke consisted of ultrafine particles (PM_{<1}).

O-18 FILTRATION OF PARTICLES OF TRAFFIC-ORIGIN ATTENUATES THEIR EFFECTS ON BLOOD PRESSURE UNDER CONTROLLED EXPOSURE CONDITIONS

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Living close to major roadways is associated with elevated blood pressure (BP) amongst other adverse health effects. There is growing evidence that ultrafine particles (UFP, <100 nm in diameter), which are elevated near major roads and highways, contribute to such risks. We assessed the efficacy of high efficiency particulate arrestance (HEPA) filtration and building envelope adjustment at reducing exposure to and health effects of air pollution next to major highways.

We used a randomized three-period crossover trial design to assign 77 participants (aged 40-75 and without diagnosis of hypertension) to three two-hour exposure sessions. Sessions were conducted in one of two rooms immediately adjacent to highways. High, medium and low exposures were attained by varying the degree of air exchange and amount of HEPA filtration in the room. During high exposure sessions, the room was ventilated with outdoor air and no filtration was used. During low exposure sessions, leakage of outdoor air into the room was minimized and HEPA filtration was maximized. During medium exposure sessions, a moderate amount of HEPA filtration was used (less than half of that used in low sessions). Indoor particle number and black carbon (BC) concentrations (i.e., markers of traffic-related air pollution) were monitored continuously.

During each session participants sat quietly and wore noise-cancelling headphones, while their BP was monitored every ten minutes using an ambulatory BP monitor. We monitored pulse and oxygen saturation continuously. Median particle number and BC concentrations for low, medium and high exposure sessions were 2,300, 9,400, and 28,800 particles/cm³ and 120, 380 and 680 ng/m³, respectively. They were relatively steady during sessions with a coefficient of variation of about $\pm 20\%$.

We used a linear mixed model, consisting of a random intercept, a Toeplitz covariance structure, and effects for exposure, period, and sequence to analyze mean changes in systolic BP. Despite systolic BP rising linearly over time during the exposure sessions ($p < 0.001$), HEPA filtration was significantly effective at minimizing changes in BP: the overall mean change in systolic BP over the course of the sessions was 0.6 mmHg during low exposure (maximum HEPA filtration), 1.2 mmHg during medium exposure (moderate HEPA filtration), and 2.5 mmHg during high exposure (no HEPA filtration) sessions (linear trend p -value = 0.019).

Our results are consistent with HEPA filtration and building envelope tightness being able to significantly reduce the effects of ambient traffic-related air pollution on BP.

O-19 CERTIFIED LABORATORY ANALYSIS OF CARBON NANOTUBE CONCENTRATIONS AS ELEMENTAL CARBON

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The U.S. National Institute for Occupational Safety and Health (NIOSH) has recommended using NIOSH Method 5040 to evaluate for exposures to carbon nanotubes by measuring for elemental carbon (EC). In this study, we compared the mass of EC quantified by NIOSH Method 5040 to the mass determined gravimetrically using carbon black and two types of multi-walled carbon nanotubes (MWCNT): small-diameter (<8 nm) and large-diameter (50-80 nm). The accuracy of this method at very low concentrations is important given a NIOSH recommended exposure limit of only 1 µg/m³ for carbon nanotubes.

A novel laboratory-based system generated an MWCNT aerosol to collect MWCNT mass ranging from 1 to 30 µg on filters. This system utilized LABVIEW software to control solenoid valves to shut off the sampling flow rate through cyclone samplers after a desired amount of aerosol mass was deposited on a filter. The system relied on real-time concentration readings from a photometer that, in combination with a known flow rate, allowed the mass collected over time to be calculated. Both quartz fiber and PVC filters were collected for assessment by Method 5040 and gravimetric analysis, respectively. Method 5040 was conducted by a certified lab and, therefore, the results obtained were considered similar to those that would be obtained by an industrial hygienist evaluating a manufacturing site. The gravimetric and EC results were compared, and a regression model was developed for each powder type. Additionally, the limit of detection (LOD) of the NIOSH Method 5040 for each powder type was determined.

The regression models had significant slopes of 0.96, 0.62, and 0.70 for carbon black, small-diameter MWCNT, and large-diameter MWCNT, respectively. Both MWCNT powder types demonstrated a statistical significant difference between the EC and gravimetric methods. Whereas, the EC and gravimetric concentrations for carbon black were not statistically different. The LOD determination resulted in values of 8.8, 4.5, and 8.3 µg for carbon black, small-diameter MWCNT, large-diameter MWCNT, respectively. Assuming a sample flow rate of 4.2 L/min and an 8-hour sample duration, the concentration-based LOD of NIOSH Method 5040 was 4.4, 2.2, 4.1, and 15.8 µg/m³ for carbon black, small-diameter MWCNT, and large-diameter MWCNT, respectively. These findings indicate the analysis of EC is affected by the structure and elemental content of the CNTs in such a way as to under-report the EC associated with the carbon nanotubes.

O-20 CHARACTERIZATION NEEDS ON NANOMATERIALS FOR REACH REGISTRATION IN 2020 - CURRENT STATUS OF SUPPORTING CHARACTERIZATION METHODS

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On 3 December 2018 the Commission adopted Commission Regulation (EU) 2018/1881 to modify REACH (Regulation for Registration, Evaluation, Authorisation and Restriction of Chemicals) Annexes I, III and VI-XII, introducing nano-specific clarifications and new provisions in the chemical safety assessment (Annex I), registration information requirements (Annex III and VI-XI) and downstream user obligations (Annex XII). The new regulation will take effect from January 1, 2020 and on essence means that nanomaterials, or chemical substances on nanoform, from January 2020 needs to be registered specifically as one or more nanoforms under each substance identity profile (SIP). The changes involve new information requirements on the specific nanomaterial characteristics. For example on the:

1. Average size or size-distribution data that should be provided to identify a nanomaterial in accordance with the European Commission proposal for definition of a nanomaterial with a threshold average minimum diameter size between 1 and 100 nm.
2. Alternatively, the volume-specific surface area (VSSA) can be used to identify the nanomaterial considering a $60 \text{ m}^2/\text{cm}^3$ a current threshold for near-spherical particles.
3. The surface coatings should be reported independent on type and level of chemical bonding.
4. The solubility and dissolution rate of NM should be reported for water and biologically relevant fluids where highly soluble NM require less new data.
5. Dustiness of powder nanomaterials should be reported to allow assessment of occupational exposure.

The data are to be applied for pure nanomaterial and substance identification, grouping, read-across and risk assessment. To reach global mutual acceptance of data, revised or new OECD Guidances Documents (GDs) and Test Guidelines (TGs) are needed to produce harmonized data on all of these physicochemical characteristics. Therefore, an urgent action, called the Malta initiative, has been taken where European countries such as Germany, France, and Denmark and the Joint Research Centre (Ispra, Italy) has taken the lead to establish different OECD GDs and TGs to enable data generation to support the new regulatory requirements. This action is supported by several EU H2020 funded projects, and in particular a dedicated work-package in the EU H2020 GOV4NANO project (<https://www.gov4nano.eu/>), and direct participation by non-European countries such as the USA, South Korea, Japan, and South Africa.

In this presentation, the new regulatory characterization requirements in REACH and actions under the Malta initiative will be introduced. The current state of art in the OECD projects on the characterization methods mentioned above will be presented along with examples of data, experimental challenges, and potential use of data.

O-21 THE CONTRIBUTION OF PERSONAL MONITORS AND SAMPLERS TO OCCUPATIONAL RISK ASSESSMENT OF ENGINEERED NANOMATERIALS

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The extraordinary physico-chemical properties of engineered nanomaterials (ENM) are currently used in numerous industrial sector in order to produce innovative consumer products. However, the mass production, widespread application and worldwide distribution of these products increased the probability for public and occupational exposure to ENMs. At the same time, several *in vitro* and *in vivo* studies showed that these xenobiotics are able to induce several toxic effects affecting different organ systems (respiratory, nervous, cardiovascular, renal, immune and endocrine systems). Consequently, there is growing concern over the adverse health effects potentially caused by ENMs when the human body takes them up.

Therefore, there is clearly a need to develop, implement and apply an adequate platform for the occupational risk assessment and management of ENMs. In this regard, the evaluation of exposure is a key stage of the complex ENM risk assessment process since, by performing environmental monitoring, it should provide important information concerning the quantity (expressed in term of particle mass, surface area or number concentration) and dimension, morphology, chemical composition and reactivity of NMs eventually present in workplaces. Indeed, to be really useful and informative in the wider context of the risk assessment/management process, an adequate sampling strategy should take into account, and then measure, all ENM metric variables that have been related to the induction of toxic effects, or at least those most relevant in exerting adverse biological responses.

Unfortunately, an international consensus on the most adequate metrics to be measured in ENM environmental monitoring has not yet been reached and it should also be noted that a comprehensive ENM characterization cannot be achieved using a single sampling instrument and/or analytical technique. For these reasons, in the past most studies investigating the ENM exposure have been carried out using either bulky static measurement equipment or not nano-specific personal samplers. However, this multifaceted approach involving the use of several sampling tools is limited by high economic cost and considerable logistic difficulties and, more importantly, it does not allow to assess individual exposure in the personal breathing zone of workers.

In this regard, innovative sampling and monitoring instruments have recently been developed and introduced to ensure an adequate evaluation of ENM personal exposure. However, considering their rather recent introduction, the number of studies that investigated their comparability and accuracy are still scarce. Nevertheless, the currently available data would seem to suggest a promising use of these personal samplers and monitors in the ENM exposure assessment. Therefore, in this context, we carried out a critical analysis of the findings obtained so far in order to identify shortcomings and gaps yet to be filled. This information will allow us to have useful indications to design and conduct further studies whose purpose is to validate the use of these instruments in the field of industrial hygiene and occupational medicine in order to have available a practical and reliable tool to assess ENM exposure.

O-22 SAFE PRODUCTION AND USE OF NANOMATERIALS IN THE CERAMIC INDUSTRY: OVERVIEW OF RESULTS FROM THE CERASAFE PROJECT

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The CERASAFE project aimed to understand nanoparticle (NP) release scenarios during industrial processes in the ceramic industry, and to assess exposure by addressing NP characterisation, release mechanisms, toxicity, and mitigation measures. The main results obtained are:

- Full physico-chemical characterisation of manufactured nanomaterials Al₂O₃, SrO, MgO, ZrO₂, Y₂O₃-ZrO₂ (YSZ) and CeO₂.
- Assessment of NP release during tile ablation and plasma spraying, evidencing that pristine and unintentional NPs may be released. NP emission mechanisms identified were nucleation, droplet ejection and shockwave ejection. Particle concentrations >10⁷/cm³ were recorded.
- Exposure assessment during ceramic tile sintering and ablation, plasma spraying, tile packaging and powdered material bagging. Particle concentrations >10⁶/cm³ were recorded. Particle concentrations modelled using one- and two-box models showed ratios modelled/measured ranging 0.89-1.03; this evidences the good performance of risk assessment models. Comparisons with risk assessment models are underway.
- NP toxicity evaluation via ALI exposure of human epithelial alveolar A549 cells for the MNMs. Two cytotoxicity endpoints were determined: plasma membrane integrity and cell metabolic activity. Unintentional NPs are being tested for cytokine measurements, DNA damage and nanoparticle aerosol uptake analyses.
- Particle hygroscopicity was used as a tool to discriminate between manufactured, unintentional and background NPs, using an HTDMA. Particle shape-factors and growth allowed for an effective discrimination between both particle types.
- Mitigation strategies were tested, showing that NP reductions of up to 85% may be achieved with proper design, implementation and maintenance. A literature review of mitigation strategies evidenced a knowledge gap regarding unintentional NPs.

Project results are uploaded to the NECID database.

O-23 OCCUPATIONAL EXPOSURE MODELLING TOOLS: REQUIREMENTS AND LESSONS LEARNT

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In the EU H2020-funded project caLIBRate (<http://www.nanocalibrate.eu/home>), different models and tools are evaluated for selection in order to support a risk governance framework for the assessment and management of human and environmental risks of manufactured nanomaterials (MNs) and MNs-enabled products. Before becoming part of the framework, the predictive capability of the tools and models need to be thoroughly analyzed and tested against observations (real data) made in actual scenarios with high quality conceptual information about MNs release and exposure.

This study highlights the specific methodology followed in caLIBRate to create an inventory of case studies with high quality data potentially available for further use for model performance testing:

1. Compilation of parameters requested by the selected models related to human exposure;
2. Identification of data sources (databases, data generated in EU Projects or literature);
3. Evaluation of data availability to cover requirements by the different models;
4. Evaluation of data quality.

More than 68 data-rich case studies have been identified and can be used for model testing. Suitability of risk assessment with different exposure assessment models ranging from simple ones applied in control banding tools (e.g. Stoffenmanager Nano v1.0 and NanoSafer v1.1β) to quantitative models (e.g. GUIDEnano) will be discussed by comparing with the gathered exposure measurement data (Figure 1).

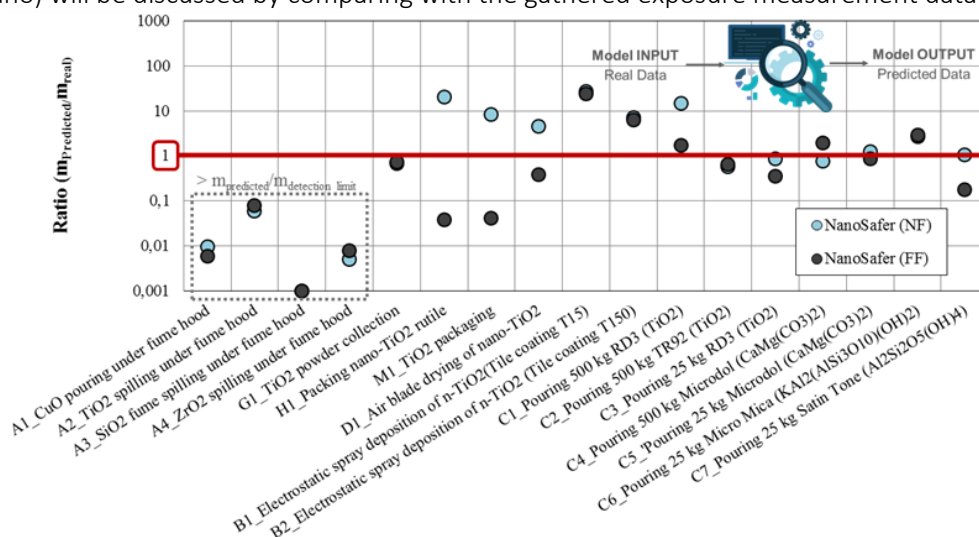


Figure 1. Example of draft model performance testing by using NanoSafer v1.1β

Acknowledgments

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O-24 TOOLS FOR RISK ASSESSMENT OF NANOMATERIALS AT THE WORKPLACE

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Despite the increasing body of toxicity data for nanomaterials, nano-specific occupational exposure limits (OELs) are still scarce, hampering the risk assessment of workplace exposure to nanomaterials. Over the past years, a wide range of (semi-)quantitative and qualitative tools have become available that can aid employers in mapping and managing possible risks of nanomaterials in their company. However, due to the large number of tools available, it has become increasingly difficult for employers to choose the most applicable tool for their own workplace situation. On request of the Dutch Ministry of Social Affairs and Employment, we have evaluated the increasing body of risk assessment and -management tools for nanomaterials. An online tool navigator was developed to support employers in finding the best available tool. The tool navigator shows approximately 40 different tools for risk assessment and -management of nanomaterials, which are evaluated for their applicability to occupational exposure in the workplace. A subset of 16 tools have been evaluated more extensively, considering e.g. input and output parameters, type of risk assessment performed, and whether the tool has been validated. In the light of this assessment, also the Dutch Provisional Nano Reference Values (NRVs) were evaluated. The NRVs, established in 2012, provide pragmatic limit values for four different classes of nanomaterials (see Table 1). They were developed based on a precautionary approach and are *not* health-based. Based on a comparison of the NRVs with recently proposed nanomaterial-specific OELs (e.g., NIOSH-RELs for carbon nanotubes and titaniumdioxide), the recent revision of the Dutch OEL for asbestos, and further development of grouping and read-across strategies for nanomaterials, it was concluded that the NRVs should be updated. Here we present the online tool navigator and a proposal to update the NRVs, including possible extension of the NRV principle for exposure to process-generated nanomaterials at the workplace and investigating the possibility of developing health-based advisory limit values for groups of nanomaterials. The updated NRVs and the online tool navigator are complementary and very useful in the risk assessment and management of nanomaterial exposure at the workplace.

Table 1: Dutch provisional Nano Reference Values (NRV)

NRV Class	Description	Examples	NRV (2012) (8-hr TWA)
1	Rigid, biopersistent nanofibres for which effects similar to those of asbestos are not excluded	SWCNT or MWCNT or metal oxide fibres for which asbestos-like effects are not excluded by manufacturer.	0.01 fibres/cm ³ (= 10,000 fibres/m ³)
2A	Biopersistent granular nanomaterial in the range of 1 and 100 nm and a density of > 6000 kg/m ³	Ag, Au, CeO ₂ , CoO, Fe, Fe ₃ O ₄ , La, Pb, Sb ₂ O ₅ , SnO ₂	20,000 particles/cm ³
2B	Biopersistent granular and fibre form nanomaterials in the range of 1 and 100 nm and a density of <6000 kg/m ³	Al ₂ O ₃ , SiO ₂ , TiN, TiO ₂ , ZnO, nanoclay, Carbon Black, C ₆₀ , dendrimers, polystyrene, Nanofibres for which asbestos-like effects are excluded	40,000 particles/cm ³
3	Non-biopersistent granular nanomaterials in the range of 1 and 100 nm	e.g. fats, common salt (NaCl)	Applicable OEL for the non-nano form

O-25 NEW INDOOR AIR QUALITY STANDARDS; ISO16000-34: STRATEGIES FOR THE MEASUREMENT OF AIRBORNE PARTICLES AND ISO16000-37: MEASUREMENT OF PM_{2.5} MASS CONCENTRATION

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Particulate matter air quality is estimated to be one of the most significant sources of adverse health effects and even very significant direct cause of death. The Global Burden of Disease (GBD) study estimated that 2.9 million deaths in 2015 were associated with household air pollution, whereas WHO estimated 4.3 million related deaths in 2012 (Lancet Commission of Pollution and Health).

Indoor air particle concentrations are a major factor for determining particle exposure and possible health risks.

The concentration, composition, and size distribution of airborne particulate matter in indoor environments can be challenging to measure reliably. Measurement strongly depend on parameters such as the room size, relative humidity, air exchange rate, air flow conditions, and sink effects on different surfaces (e.g., walls, ceilings, floor coverings, furnishings). Depending on conditions this can result in highly variable levels of indoor PM pollution that are not easily determined or measured in terms of their impact on health.

Increased need for reliable and comparable measurements has brought up a requirement for the standardisation of measurement of fine and ultrafine (PM₁₀, PM_{2.5} and UFP) particles and their real-time behaviour in indoor air. This work has been carried in ISO Technical Committee ISO/TC 146, Air quality, Subcommittee SC 6, Indoor air. The newly published ISO 16000-34 and 37 standards include for the first time a comprehensive guidance on indoor air quality airborne particle measurement instrumentation and methods. ISO16000-37 presents an EN12341 based method for measuring indoor PM_{2.5} together with supplementary real-time measurement.

This presentation will review the new ISO 16000-34 and 37 standards, evaluate their meaning for the indoor air exposure studies and give insights and instrument solutions providing both PM_{2.5} and real-time data.

O-26 OLFACTORY AND CENTRAL NEUROTOXICITY OF OCCUPATIONALLY-RELEVANT PARTICULATE AEROSOLS AND NANOMATERIALS

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Particulate aerosols generated at the workplace can be inhaled and often deposit in the nose and/or lung. Agents deposited in the nose can reach the brain via the olfactory sensory neurons (OSNs) that are normally associated with perception of smell. OSNs extend into the brain from the air interface in the nose and have direct access to odorant molecules, as well as allergens, airborne pollutants, toxic chemicals and microorganisms. Some toxicants, including chemical, metallic and non-metallic particulate compounds, damage the olfactory region leading to olfactory dysfunction that is manifested as a reduced sense of smell. Indeed, environmental air pollutants can cause nasal pathology, neuroinflammation, blood-brain barrier disruption, and Alzheimer's disease (AD)-like pathology in humans and animals. Loss of olfaction frequently precedes the hallmark clinical signs of neurodegeneration seen in AD and Parkinson's disease. Similarly, the olfactory region is often the first site to display Lewy bodies, amyloid deposits and neurofibrillary tangles that are hallmarks of classic neurodegenerative diseases. Recent experimental data suggest that olfactory and central neurotoxicity is elicited by a variety of occupational particulates (welding fume, fracking sand dust) and engineered nanomaterials (multi-walled carbon nanotubes, silver nanoparticles), raising concerns of potential long-term neurological health risks linked to such particulates. The translational relevance to humans depends upon the interspecies anatomical and structural differences in nasal architecture, toxicokinetic modeling, as well as, epidemiological studies to assess injury/disease outcome and risks.

Disclaimer: The findings and conclusions in this report are those of the author(s) and do not necessarily represent the official position of the National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention.

O-27 PARTICLE CELL INTERACTIONS IN THE ALVEOLAR REGION OF THE LUNGS AND THEIR RELEVANCE FOR ASSOCIATED HEALTH EFFECTS

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It is well known, that depending on their aerodynamic size, inhaled particles may penetrate deep into the respiratory area of the lung. Particularly nanoparticles are known to deposit efficiently in the most vulnerable, alveolar region, the epithelial cell sheet which is not protected by a thick mucus layer or mucociliary clearance. Phagocytic clearance, realized by specialized alveolar macrophages (AM), is therefore the major route of particle removal from the alveolar surface. According to our current understanding, AMs originate developmentally from fetal monocytes, which after birth differentiate into long-lived cells. They proliferate slowly and leave their alveolar residence only sporadically by finally getting cleared via the mucociliary escalator. Because of this slow removal also of particle laden AMs, the physiological cleansing function works most effective for biodegradable particles, like organic dust, cell debris or even many pathogens, but reaches its limits for inorganic and biopersistent nanoparticles. Since AMs interact rapidly with inhaled particles and may contain internalized materials over a long period of time, they are among the first and primary cell types that process nanoparticles and mediate the innate host responses in a material dependent manner. Particles cytotoxic to phagocytes may even cause a depletion of the originally resident AM pool, which will be dynamically substituted by blood monocyte derived cells. At present it is not clear to which extend this new cell population might differ in its capability to maintain the integrity of the alveolus.

I will show evidence that pulmonary deposition of high surface area nanoparticles irritates the alveolar epithelium and thereby cause a rapid inflammatory response, characterized by the airspace accumulation of neutrophilic granulocytes. For particles of low toxicity, this inflammatory response can resolve within a few days after exposure, while for materials of persistent toxicity to macrophages, conditions of chronic inflammation can develop. I will discuss the hypothesis, that even though the foreign matter may get rapidly phagocytized by AMs, dependent on the material toxicity, the particulate hazard can (i) either be kept safe by the phagocytes allowing a rapid resolution of inflammation, or (ii) in case of biopersistent, macrophage-toxic materials, causes a sustained inflammatory stimulation of the epithelium since impaired macrophage clearance of remaining matter leads to continuous inflammatory cell recruitment with defective resolution.

O-28 EVALUATION OF PARTICLE MASS, NUMBER AND VOLUME SPECIFIC SURFACE AREA AS DOSE METRICS FOR INTERNALIZATION OF NANOPARTICLES

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The extent of uptake of nanoparticles (NPs) by biota is commonly expressed on a mass basis. There are indications, however, that this does not provide an accurate picture of NPs internalization. Other dose metrics such as particles number and volume specific surface area (VSSA) may be more suited to properly express the effective dose, and will allow for a basically better understanding of NPs uptake. Up till now, investigations on a cell-by-cell basis to evaluate different dose metrics for NPs uptake and toxicity has been challenging due to limitations in analytical techniques. In this study, we used the recently developed technique of single cell Inductively Coupled Plasma Mass Spectrometry, to systematically study the uptake of gold NPs by algal cells at the cellular level, comparing particle mass, particle number and VSSA as possible dose metrics suited for quantifying NP uptake. The aim is to elucidate how particle size and shape affect particle internalization as expressed by different dose metrics. Our finding showed that the NPs are not taken up equally across cells within the algae population. Some cells internalize NPs in numbers of up to hundreds particles per cell and some cells do not internalize particles at all. Our findings elucidated that particles number followed by VSSA were more suited dose metrics than particle mass to quantify the uptake of NPs. We observed linear relationships between particle internalization and the particle number in the exposure media when the particles number is considered as the dose metric, regardless of shape and size of the particles. Hence, a key finding of our study is that, opposed to common belief there is no selectivity of particle uptake by cells irrespective of particle size and shape and uptake is proportional to the effective number of particles to which the cells are exposed to.

O-29 RESOLVING MOLECULAR EVENTS BY SUPER-RESOLUTION MICROSCOPY EXEMPLIFIED ON INTERACTION OF TiO₂ NANOTUBES WITH CELL LAYERS IN LIVE

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Recently, advanced microscopic techniques have enabled the discovery of the lipid wrapping soon after the exposure of living lung epithelium to TiO₂ nanotubes. Strong affinity of TiO₂ surface towards lipid head groups drive this phenomenon of wrapping TiO₂ nanomaterials such as nanotubes by pieces of cell membranes (*Nano Lett.*, **2018**, 18 (8), pp 5294–5305). Proteomics, which has been performed on these wraps to unravel their protein composition, identified more than 500 different proteins not present in controls and suggesting interference with various signalling cascades such as coagulation cascade.

Recently, our further studies, in which we track supramolecular rearrangement with STED super-resolution microscopy in live, revealed interesting degradation phenomena triggered by the presence of TiO₂ nanotubes. Namely, the aggregation of lysosomes was observed as well as the creation of new composite materials inside the living cells in a form of composite of nanoparticles and membranes as well as of composite of nanoparticles and actin fibres (see Figure 1).

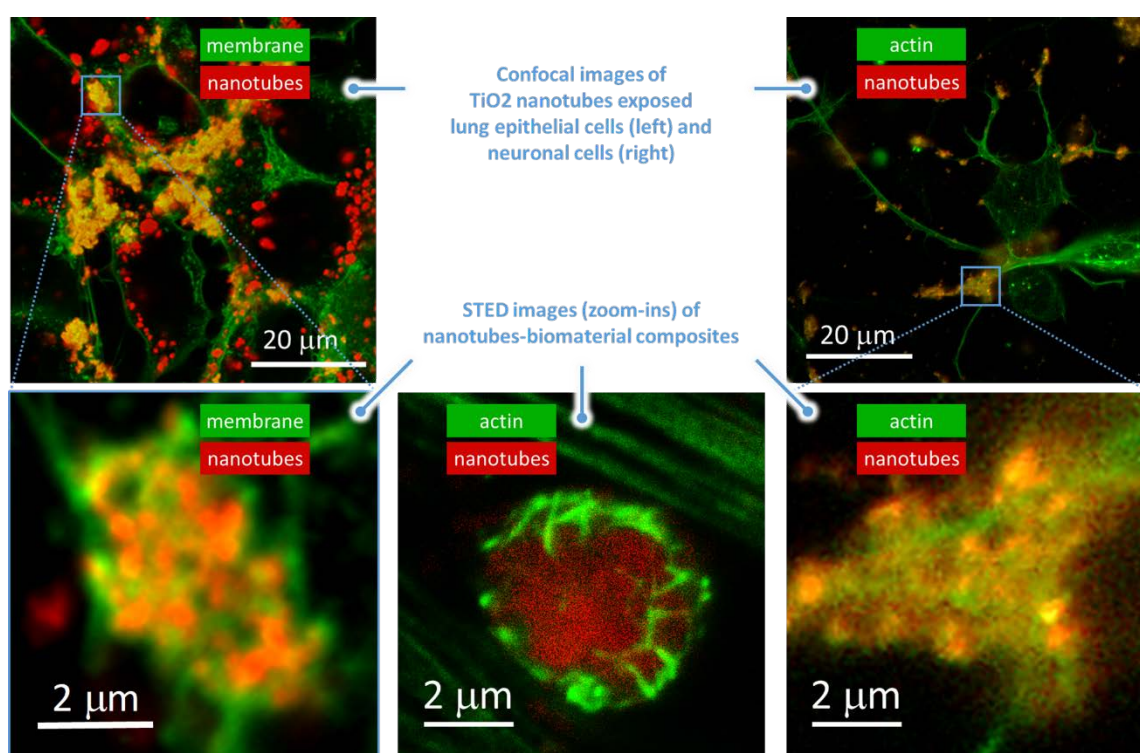


Figure 1: Confocal fluorescence images of living cells exposed to TiO₂ nanotubes and their STED zoom-ins showing appearance of new composite structures composed of nanotubes and (supra)molecular structure of biological origin such as membranes and actin fibres.

O-30 NANOMATERIALS ACCELERATE SYSTEMIC AUTOIMMUNE DISEASE IN LUPUS-PRONE NEW ZEALAND MIXED MICE

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The majority of research on the potential adverse effects of nanomaterials has focused on lung inflammation. However, little is known regarding systemic effects of nanomaterials. We have previously reported that crystalline silica, known to cause lung inflammation and autoimmune disease in people, exacerbates systemic autoimmune disease in various autoimmune prone murine models. Therefore, we examined whether a number of high volume engineered nanomaterials can accelerate systemic inflammation in NZM2410 autoimmune prone mice. Both male and female NZM2410 mice (9-11 weeks of age) were given 50 µg/mouse (4 different MWCNT, NiO, TiO₂) or 1 mg/mouse of silica as a positive control or dispersion medium as the vehicle control. The dosing was once/week for 4 weeks and the mice harvested 7 weeks after the last instillation. Various tissues and plasma were collected and analyzed for evidence of inflammation and systemic disease. Only NiO and SiO₂ caused elevated lung lavage cell counts and statistically significant elevation of IL-1β release by isolated lung macrophages *ex vivo*. There was no consistent pattern of significant changes in plasma cytokines. SiO₂ and Mitsui-7 MWCNT caused elevations of IFN-γ, IL-33 and IL-6 and SiO₂ also elevated IL-10, TNF-α and IL-1β in lung lavage fluid. Antinuclear antibodies were elevated in plasma of mice treated with Mitsui-7, NiO or SiO₂. Flow cytometry analysis of lung and spleen tissue demonstrated that both SiO₂ and Mitsui-7 decreased B cells (CD19+) in lung tissue and also blood. SiO₂, Mitsui-7 and another MWCNT significantly decreased total AM in the lung, but increased Class II+ inflammatory macrophages in the lung. There were no major differences in response of the mice based on sex. Taken together, the results suggest that in addition to SiO₂, both NiO and Mitsui-7 can cause systemic disease in the NZM2410 murine model and the ENM can be transported to lymphoid tissues from the lung. The work was supported by NIH grants P30 GM103338 and ES021464.

O-31 MWCNT INHALATION TOXICITY – INFLUENCE OF AEROSOL CHARACTERISTICS

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Carbon nanotubes (CNTs) are graphene layers in a cylindrical form being used in an increasingly wide range of applications. The growing production of CNTs has increased their potential for impacting on human health, especially for occupational exposures and biomedical applications such as drug delivery, biosensors, tissue engineering etc. Animal studies have indicated that pulmonary exposure to CNTs is linked to a series of adverse respiratory effects, including lung inflammation, genotoxicity, fibrosis and granuloma formation, the degree and characteristics of which are dependent upon the detailed physico-chemical properties of the material that is inhaled. To be able to assess the effect of variations in physico-chemical properties on both acute and chronic pulmonary effects, two different multi-walled CNTs (MWCNTs) with well-characterized physico-chemical properties were tested for effects on respiratory function in animals. Included was a type of pristine MWCNT (pMWCNT) and a chemically functionalized MWCNT (MWCNT-COOH).

The tested pMWCNT is a long ($4 \pm 2 \mu\text{m}$) and thick ($70 \pm 20 \text{ nm}$) MWCNT, and exists as single fibers, tangled clumps of fibers and loose, generally spherical, agglomerates after aerosolization, whereas the second fibre tested MWCNT-COOH is a very long ($10 - 30 \mu\text{m}$) and thin ($< 8 \text{ nm}$) MWCNT, and primarily formed much denser sphere-like aerosol agglomerates. Sprague Dawley rats were exposed to both aerosols produced using an acoustic generator in a whole-body chamber at two exposure concentrations for each material (range 0.5 to 4.5 mg/m^3), for 6 hours/day, 5 days/week for 4 weeks. Controls were exposed to filtered air. Groups were sacrificed at 3 days, 30 days and 1 year. Large numbers of MWCNTs were observed forming agglomerates inside bronchoalveolar lavage fluid (BALF) macrophages which reduced/cleared over time. Results from analysis of BALF lung histopathology and global mRNA expression in lung tissue indicate that both aerosolized MWCNTs produced limited toxicological effects but with some different characteristics in each case. RNA-SEQ analysis results from the above MWCNT inhalation studies have been compared to those from previous inhalation studies of other nanoparticles (Ag nanoparticles and CeO_2 nanoparticles) and the correlation with nanomaterial aerosol characteristics is discussed.

O-32 IMPORTANT INFERENCES FROM ANIMAL EXPERIMENTS ASSESSING ADVERSE HEALTH EFFECTS OF METAL OXIDE NANOPARTICLES

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Abstract. Nanoparticles (NPs) of Fe₃O₄ were produced in a chemical technique and nanoparticles of Ag, Au, CuO, NiO, Mn₃O₄, PbO, ZnO, TiO₂, SiO₂, Al₂O₃ - through laser ablation. In some experiments, we compared particles of a given chemical composition having different diameters, while in others – equidimensional nanoparticles of different metals or metal oxides (Me-NPs). We used two experimental models: a single intra-tracheal instillation of Me-NPs 24 h before the bronchoalveolar lavage procedure (collecting) and the repeated intra-peritoneal injections during 6-7 weeks in non-lethal doses. Besides, we carried out long-term inhalation experiments with NPs of Fe₂O₃, SiO₂ and NiO.

We have demonstrated that NPs are much more noxious as compared to their fine micrometric or even submicron counterparts and are usually the more toxic the smaller their dimensions within the nano-scale range. We found also that toxicity of Me-NPs strongly depends on their chemical nature, solubility and mechanisms of action specific for a given metal in any chemical form. Solubilization of Me-NPs in biological milieus plays an important part in their toxicokinetics, which can prevail over that of the physiological mechanisms controlling their distribution, retention and elimination. On the other hand, thanks to the high activity of these mechanisms, the organism is not defenseless against the impact of Me-NPs.

The nonspecific responses of the organism to the impact of Me-NP included: changes in the cytological and some biochemical characteristics of the bronchoalveolar lavage fluid caused by the deposition of particles in the lower airways; various manifestations of systemic toxicity, particularly expressed damage to the liver and kidneys; some cytological signs of a likely onset of hyperergic inflammation; moderate neurological disturbances presumably associated with possible penetration of Me-NP into the brain from the blood (in which its concentration rises as a result of proven solubilization of metal oxide nanoparticles in biological media), and with the transport of nanoparticles from the nasal mucous membrane along the olfactory pathway; a paradoxically low manifestation of pulmonary pathology explained by low chronic retention of nanoparticles in the lungs; a genotoxic effect on the organism level even under low chronic exposure for which systemic toxicity is mild enough.

As a protective measure, the toxicity and even genotoxicity of Me-NPs can be significantly attenuated by adequately composed combinations of some bioactive agents in innocuous doses.

O-33 CHARACTERIZATION OF NANOPARTICLES IN FOOD AND BIOLOGICAL MATRICES BY ICP-MS BASED METHODS

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With the increasing use of nanotechnology, there is a need for reliable detection and characterization methods for nanoparticles (NPs) in food and biological matrices. Inductively coupled plasma-mass spectrometry in single particle mode (spICP-MS) has become a frequently used technique during the last few years. SpICP-MS has been applied in our laboratory for studying inorganic nanoparticles in a variety of biological samples, including rat lung and liver tissue (Au and CeO₂ NPs), whale brain and liver tissue (HgSe NPs), human synovial fluid (Co- and Cr-containing NPs) and human placenta tissue (Ag and TiO₂ NPs). Furthermore, food-related samples were investigated including lean chicken meat (silver NPs), game meat (lead NPs), food simulants (Ag NPs), noodles (Al-containing NPs) and candy (titanium dioxide NPs).

Our experiences show that spICP-MS is a powerful screening method for the presence of NPs, but that care has to be taken with regards to false-positive-results and the obtained quantitative information in terms of particle size distribution (PSD) and number / mass concentration. The relatively easy implementation of spICP-MS in state-of-the-art ICP-MS instruments (which can be otherwise used for metal analysis and speciation) makes it a promising technique for routine analysis despite the analytical limitations.

Another often-applied technique is asymmetric flow field flow fractionation (AF⁴) hyphenated with ICP-MS. A number of examples where AF⁴-ICP-MS was applied are Ag NPs in chicken meat, SiO₂ NPs in tomato soup, Co- and Cr-containing NPs in human synovial fluid, Al₂O₃ and TiO₂ particles in toothpaste as well as nanoplastics spiked to fish. Our studies showed that AF⁴-ICP-MS can be very helpful for characterizing complex samples and for obtaining reliable mass-based PSDs but it is not an ideal method for determining the number-based PSD. In order to acquire accurate data the AF⁴ separation method and settings must be optimized for each new sample matrix and NP combination.

Electron microscopy is always required as confirmative technique and the only technique that can provide information on particle shape and distinguish primary particles and particle aggregates/agglomerates.

We identified sample preparation as a crucial step, especially in the case of solid/semi-solid matrices where simple dilution is not sufficient. As spICP-MS analysis is not as sensitive to eventually remaining matrix residues as AF⁴, complete digestion of the matrix is usually not required. The main challenge is to minimize changes of the NPs during sample preparation mainly due to dissolution. For the majority of examples, we identified enzymatic digestion as the most suitable sample preparation method. Based on our experiences, the talk will highlight the possibilities and limitations of the before mentioned techniques and potential future developments.

P-1 TOXICITY EVALUATION OF AIRBORNE CERAMIC THERMAL SPRAYING-GENERATED NANOPARTICLES IN A 3D HUMAN BRONCHIAL EPITHELIAL MODEL

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There is considerable evidence that air particulate matter (PM) adversely affects human health and nanosized particles are expected to play a major role. High Velocity Oxy-Fuel (HVOF) spraying, a commonly used thermal technique for application of ceramic coatings onto metallic surfaces, has a high potential for nanoparticle release into the workplace environment that raise occupational health concerns. This study aimed to investigate the toxicity of airborne nanoparticles released during HVOF in *MucilAir™* human 3D bronchial epithelium from a 30-year-old donor under air-liquid interface (ALI) conditions. The airborne particulate matter (PM) < 2.5 µm and ultrafine (UFP) < 0.2 µm fractions were sampled at a ceramic industrial facility using an aerosol concentration enrichment system (VACES). *MucilAir™* cultures were exposed three consecutive days to different deposited doses of the aerosolized PM 2.5 (1033-3099 ng/cm² per day) or UFP (560-1680 ng/cm² per day) fractions in a Vitrocell® Cloud 12 system. Cytotoxicity was evaluated by the lactate dehydrogenase (LDH) release (24h after each exposure) and WST-1 metabolization (24 h after the last exposure) assays. Primary and oxidative DNA damage were evaluated in cultures collected 24 h after the last exposure by the alkaline and FPG-modified comet assay versions, respectively. The chemical analysis revealed that airborne HVOF-generated nanoparticles were mainly constituted by WC, CrC and Ni. Statistical analyses were carried out from TEM images acquired on grids exposed to aerosols in the same manner as the cells. TEM analyses revealed a polydisperse index of 0.28 for UFP fraction at a concentration of ~2x500ng/cm², while for the PM fraction at a concentration of 2x1µg/cm² the polydisperse index was calculated to be 0.47. It is also worth mentioning that EDS elemental analyses reveal a significant presence of Ag in the aerosolized samples. Exposure to the aerosolized PM 2.5 fraction failed to affect plasma membrane integrity of the *MucilAir™* cultures as evaluated by the LDH release. However, 24h after the last exposure a significant decrease in cellular metabolic activity in human bronchial epithelium exposed cultures compared to the control cultures was observed, as assessed by the WST-1 assay. On the other hand, cultures exposed to the lowest tested dose of the aerosolised UFP fraction exhibited a significant increase in LDH release only visible at 24 h after the first exposure, whereas no changes in the metabolic activity were detected. On the other hand, only human bronchial cultures exposed to the highest tested dose of the UFP fraction aerosol exhibited a significant increase of oxidative DNA damage comparing with control cultures. Thus, our findings highlight the potential health risks associated with exposure to unintentional emissions derived from ceramic industry processes.

This work was supported by the Portuguese Foundation for Science and Technology (FCT) through the ERA-NET SIINN project CERASAFE (SIINN/0004/2014).

P-2 INDOOR AND OUTDOOR PARTICLE NUMBER CONCENTRATION IN THE SAPIENZA UNIVERSITY CAMPUS OF ROME

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In the context of VIEPI project (Integrated Evaluation of Indoor Particulate Exposure), the evaluation of Indoor and Outdoor Particle Number Concentration (PNC, a proxy of Ultrafine Particles) was carried out in the Fermi Physics building of Sapienza University in Rome, through three monitoring campaigns realized during the autumn 2017 and 2018 and the spring 2018.

The object of this study was to assess exposure of people to PNC in two seasonal periods characterized by different meteorological conditions and different use conditions of indoor environments. PNC measurements were carried out with high spatial and temporal resolution, using Condensation Particles Counters (CPC3007, TSI), at 1 minute resolution, in five classrooms used for teaching activities in two time slots (from 8:00 to 12:00; from 14:00 to 18:00). In addition, measurements of temporal variability of size distribution from 10 nm to 10 μm , were carried out with a Particle Sizer Spectrometer 3910 TSI and two OPS 3330 TSI. Each monitoring campaign was realized for two weeks. During the first one, to assess PNC on the same level, indoor measurements were carried out in classrooms located on the second floor but with different orientations (Classroom 3 facing the north side, Classroom 4 facing the south side, Computer Lab facing the east side). In the second week, to evaluate PNC at different heights, indoor measurements were made in classrooms located at different levels: one classroom (Nicola Cabibbo conference hall) on the ground floor, Computer Lab on second floor and Classroom 7 on the fourth floor. For each campaign, in order to assess the time variation of indoor/outdoor ratios, indoor measurements were evaluated in relation to outdoor concentrations (Outdoor A and Outdoor B) measured at the same time and on the same level during the first week and on different levels in the second week. During the two seasonal samplings, information about the indoor environment were collected: number and time of open windows and open doors, air conditioners or heating systems on or off, number of occupants, performed activities (lectures, exams and seminars). The monitoring campaign highlights that the indoor concentrations were generally lower than outdoor concentrations in winter season, while, in the summer, the indoor PNC were higher or comparable to the outdoor particles. In figure 1, the indoor and outdoor PNC trends registered on a spring day and on an autumn day in the Classrooms 3 and in Classroom 4, are showed.

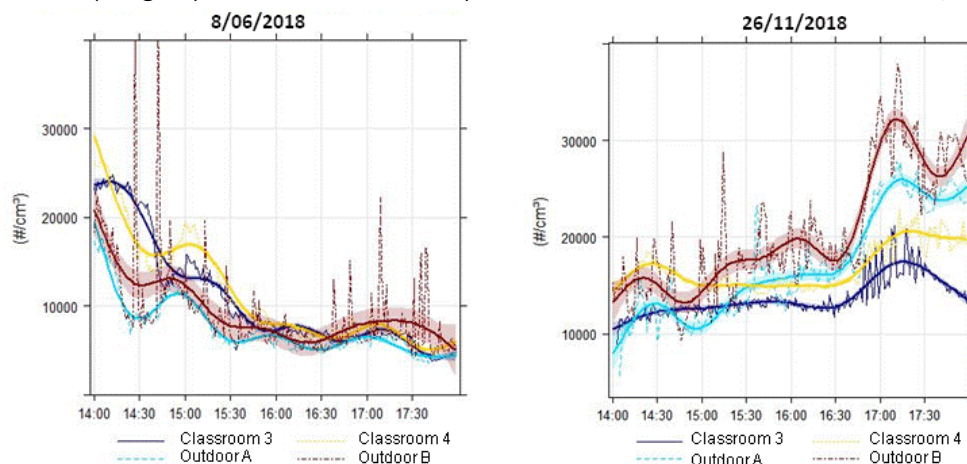


Fig. 1: Outdoor and Indoor PNC on a spring day and on an autumn day

P-3 ROAD-MOTOR TRAFFIC COMPLEX AS A SOURCE OF POLYAROMATIC HYDROCARBONS CONTAINED IN FINE DUST PARTICLES

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Road-motor traffic complex (RMTC) is a transport system of every urban settlement. Exhaust gases include: CO, NO_x, non-methane volatile organic compounds, greenhouse gas, oxidizing agents, particulate matter (heavy metal compounds), persistent organic pollutants, toxic substances (dioxins and furans). Carcinogenic substances based on polyaromatic hydrocarbons (PAHs) are significant.

When hot exhaust gases are cooled during RMTC operation, atmospheric polyaromatic hydrocarbons produce condensation aerosols. Moreover, fine soot particles PM_{2,5} are an excellent adsorbent for PAHs. In cities, daily variations of PAH concentrations follow traffic intensity dynamics (WHO, 2001).

Chromatographic study of atmospheric air samples in the vicinity of a highway with traffic flow rates of 2500–3000 vehicles/hour made it possible to identify a wide range of PAHs. The following chemicals have been detected: benzo(a)antrecene, benzo(a)pyrene, benzo(e)pyrene, benzo (b)fluoranthene, benzo(k)fluoranthene, benzo(ghi)perylene, dibenz(a, h)anthracene, coronene, pyrene, chrysene. Three of them (dibenz(a,h)anthracene, benz(b)fluoranthene and chrysene) manifest a pronounced carcinogenic effect.

The air pollutant emission inventory guidebook EMEP/EEA allows to estimate the amount of PAH emitted into atmospheric air during fuel combustion in the process of RTMC operation. This information is not currently taken into account in all countries during motor traffic emission inventory.

Results of calculations carried out for the megapolis where car fleet is under 2 million vehicles, and the level of motorization is within 300 cars per 1000 residents showed that fuel combustion in the process of RTMC operation results in release of 49 tons of highly toxic polycyclic aromatic hydrocarbons into the atmospheric air. Maximum allowable concentrations for PAH are mainly measured in micrograms per m³. It indicates the need to determine the level of carcinogenic risk to public health.

It has been revealed that the level of individual carcinogenic risk for population of the central part of the city under study, having 0.5 million residents, is 6×10^{-2} , which according to international criteria requires organizing and carrying out emergency measures to reduce it.

Thus, when studying the chemical composition of PM_{2,5}, released into atmospheric air of populated areas during RTMC operation, PAHs manifesting pronounced carcinogenic effect (benzo(a)pyrene, dibenz (a,h)anthracene, benzo(b)fluoranthene and chrysene) were identified.

Hence, when organizing monitoring of fine particulate matter in atmospheric air of residential areas it is necessary to assess chemical composition, in particular of PAHs, as well as its amount.

P-4 FORMATION OF ULTRAFINE PARTICLES IN A CLASSROOM UNDER DIFFERENT VENTILATION CONDITIONS

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An important contribution of human exposure to particles occurs indoors, since people spend a major part of their days indoors, at home, at work or at school. However, there is little knowledge on particle characteristics and sources indoors. Our study focuses on submicron particles formed under real-life conditions in a primary school.

Particle size distributions (in the range 10 – 400 nm) were measured in a classroom as part of a project to optimise air quality and the use of energy for ventilation. The school was built recently and opened in autumn 2016. It is equipped with low emitting interior and a demand controlled ventilation system. The measurements were carried out continuously (1 minute time resolution) for 3 weeks in May/June 2018 while the classroom was in normal use. A full class had 24 pupils aged 9-10 years and 1-2 teachers. Also measurements of volatile organic compounds, O₃, NO₂, CO₂ and ventilation rates in the room were available. Different ventilation strategies were tested during the field campaign.

Particle formation and growth was observed frequently and could often be related to changes of the ventilation rate. The formation of ultrafine indoor particles occurs through reactions between ozone and volatile organic compounds emitted by occupants themselves, paints, building products and by furniture. Activities, such as peeling an orange in the presence of ozone, may also result in new particle formation. The first particle formation event of the day was generally observed in the mornings after the ventilation was turned on and ozone from outdoors was brought into the classroom where it reacted with VOCs from indoor sources. The events usually lasted for 2 hours and were often still ongoing when the pupils entered the class room at 08:30. In the afternoon, after the ventilation was turned off, the ozone present in the room was consumed by VOCs. There were also particle formation events during the school day, presumably caused by the presence of occupants and their activities.

We will present the contributions from different VOCs to the formation of secondary organic aerosol and identify the main sources leading to particle formation in the classroom.

P-5 NON-INVASIVE SENSITIVE METHOD FOR OXIDATIVE STRESS QUANTIFICATION IN EXHALED AIR

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Inhalation exposure to xenobiotics like certain chemicals agents or particles has a recognized impact on the development of lung diseases such as chronic obstructive pulmonary disease (COPD). Although in more than 80% of cases smoking is the main etiological factor in COPD, other factors are also implicated, such as indoor or atmospheric air pollution, certain genetic factors or industrial activities.

In particular, Inhalation of particulate matter (PM) is known to induce via an immune mechanism, the production of reactive oxygen species (ROS) that in turn can lead to oxidative stress, inflammation and ultimately increase the risk of disease. The diagnosis and assessment of COPD currently relies on respiratory functional exploration test using spirometry. However, this technique does not allow early diagnosis nor early detection of exacerbation episodes. Therefore, in addition to spirometry, alternative methods focusing on exhaled air as non-invasive matrix are being developed in the ROBoCop* project in the framework of CliniMARK EU-COST Action (CA16113) in order to access the biochemical information related to lung oxidative stress.

One studied approach focuses on the simultaneous quantification of a panel of tree oxidative stress biomarkers in exhaled breath condensate (EBC) samples by LC-MS/MS technique: 8-hydroxy-deoxyguanosine, 8-isoprostaglandin F2 α and malondialdehyde. Providing that EBC is a strongly diluted sample, the main challenge consists of reaching low limit-of-quantification as well as robust sample pre-concentration step.

In parallel, we aim at optimizing a portable photonic system, the OPEA analyzer [1], able to determine lung oxidative potential via direct analysis of exhaled air (1L). The detection principle relies on multiscattering-enhanced absorbance strategy that enables fast and sensitive determination of the homeostatic redox balance if lining fluid droplets contained in the exhaled breath aerosol. Optimization aspects related to instrument design and sample storage will be addressed.

Both biomarker quantification approaches will be discussed in terms of analytical validation and expected context-of-use in the COPD paradigm.

**Respiratory disease Occupational Biomonitoring Collaborative Project*

[1] Laulagnet, A., et al. (2015). "Sensitive Photonic System to Measure Oxidative Potential of Airborne Nanoparticles and ROS Levels in Exhaled Air." *Procedia Engineering* 120: 632-636.

P-6 STUDY FINDINGS OF CHEMICAL COMPOSITION OF FINE DUST PARTICLES IN THE AREA OF ROAD-MOTOR TRAFFIC COMPLEX EXPOSURE

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For better insight into the components of the system under study, the term “road-motor traffic complex” (RMTC) is used, which is a combination of traffic flows of varying intensity along the urban road network and the road network itself. In this case, the levels of motorization among the residents and the degree of road network development of the area are considered to be the indicators of RMTC development intensity.

The annual average PM_{10} concentrations in atmospheric air of the areas in the vicinity of major highways increase by approximately 6% per year. This trend is associated with an increase in the number of vehicles and the intensity of their operation. The average annual PM_{10} concentration near major highways currently ranges between 40 and 49 $\mu\text{g}/\text{m}^3$. (Huhtala, M., 1990). Studies of total amount and chemical composition of suspended particulate matter (TSP) and fine dust particles (PM_{10} and $PM_{2.5}$) as air pollution factors caused by fuel combustion and operational wear of RMTC are sparse.

Studies were conducted in residential areas along urban highways with varying motor traffic intensity. The following classification is assumed: a highway with traffic intensity of 2500–3500 vehicles/hour; 1500–2000 vehicles/hour and ≤ 500 vehicles/hour.

Maximum TSP concentrations along the highway with traffic intensity of 2500 vehicles per hour reached 13.5 mg/m^3 . According to chemical analysis findings 70% of the TSP in studied atmospheric air samples were found to be aluminium silicate microscopic particles.

Table. MAC exceeding degree of airborne particulate matter in residential areas with varying traffic intensity

Particulate matter	MAC (single), mg/m^3	MAC exceeding degree (Concentration/MAC) with varying traffic intensity		
		≤ 500 (vehicles/hour) (number of samples $n = 112$)	1500–2000 (vehicles/hour) (number of samples $n = 128$)	2500–3000 (vehicles/hour) (number of samples $n = 112$)
TSP	0.5	0.61	2.1	2.86
PM_{10}	0.30	0.39	1.49	1.98
$PM_{2.5}$	0.16	0.15	0.51	0.91

The level of air pollution (TSP, PM_{10} and $PM_{2.5}$) in areas with intensive traffic flow is affected by the process of primary dust formation (roadway surface and vehicle component destruction) as well as by fuel combustion and secondary dust formation.

In the study of the chemical composition of TSP, sampling was carried out continuously for 10–12 hours in the daytime. Studies have shown that average concentrations of zinc, iron, cobalt, lead, chromium, and nickel exceed hygienic standards even at a traffic intensity of over 1 500 vehicles/hour. Thus, it was revealed that highway traffic results in generation of TSP, PM_{10} , $PM_{2.5}$ and formation of turbulent air flows causing secondary entrainment of particulate matter particles containing a complex of heavy metal compounds in atmospheric surface layers, in addition to combustion products release into atmosphere.

P-7 CHARACTERISATION OF AIR CONTAMINANTS EMITTED DURING LASER CUTTING OF CARBON FIBER REINFORCED COMPOSITE MATERIALS

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Fiber reinforced polymer composite materials (CFRP) are used in increasing quantities in a large number of technologies such as aviation, automotive, building & construction, electronics and sport & leisure owing to their high strength-to-weight ratio and excellent fatigue resistance. Three basic types of fiber materials are commonly in use in the composite industry; carbon/graphite, aramid and glass fibers. Among the different fibrous materials, carbon fibers have special capacity to retain their tensile strength at high temperature independent of moisture. Since the thermal decomposition and degradation are varied, a detailed investigation of emissions during laser cutting of different composite materials is required to develop protective environmental and occupational measures as well as production strategies for adequate process management.

The emission of ultrafine carbonaceous particles during the laser cutting of CFRP was investigated. The study was based on characterisation of air contaminants emitted during laser cutting of an epoxy based CFRP material with respect to particle size distribution, particle morphology and chemical composition. Results indicate that about 90% of the total particulate mass is present as fine particulate matter with an aerodynamic cut-off diameter of 0.25 μm , considerable amounts of ultrafine carbonaceous particulate matter dominated by organic carbon are emitted during high power laser cutting of CRFP.

P-8 PROCESSING OF NANOCOMPOSITES AND GROUPING OF EXPOSURE

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In the course of the German project nanoGRAVUR to develop a framework for the grouping of nanomaterials concerning their risks, the IFA performed the determination of exposure during grinding of selected nanocomposites. The goal was to investigate whether grouping of physical-chemical characteristics of nanomaterials influences the exposure and thus the risk. Composites on basis of epoxy resin, hardened cement mixtures, and aluminium with the additives of nano objects from the groups of fibres (CNT, WS₂), platelets (graphene) and compact particles (TiO₂, SiO₂, carbon black) were chosen. Grinding exposure tests were carried out in a ventilated test chamber (4 m², 10 m³, 30 m³/h). Number and mass concentrations were determined at the worker and near to him. Particle number concentration ranged between 5600 and 23000 1/cm³ and only the epoxy resin composites showed a decrease with adding nano objects, lowest with WS₂. The respirable dust concentrations ranged from 0.6 to 46 mg/m³ in this specific setup, highest for cement composites, lowest for aluminium composites. Normalising the concentrations to the area ground on the composite pieces reveals the ranking given in the figure below. The added nanomaterial is less influencing the exposure than the matrix material, which is in accordance to pure release measurements.

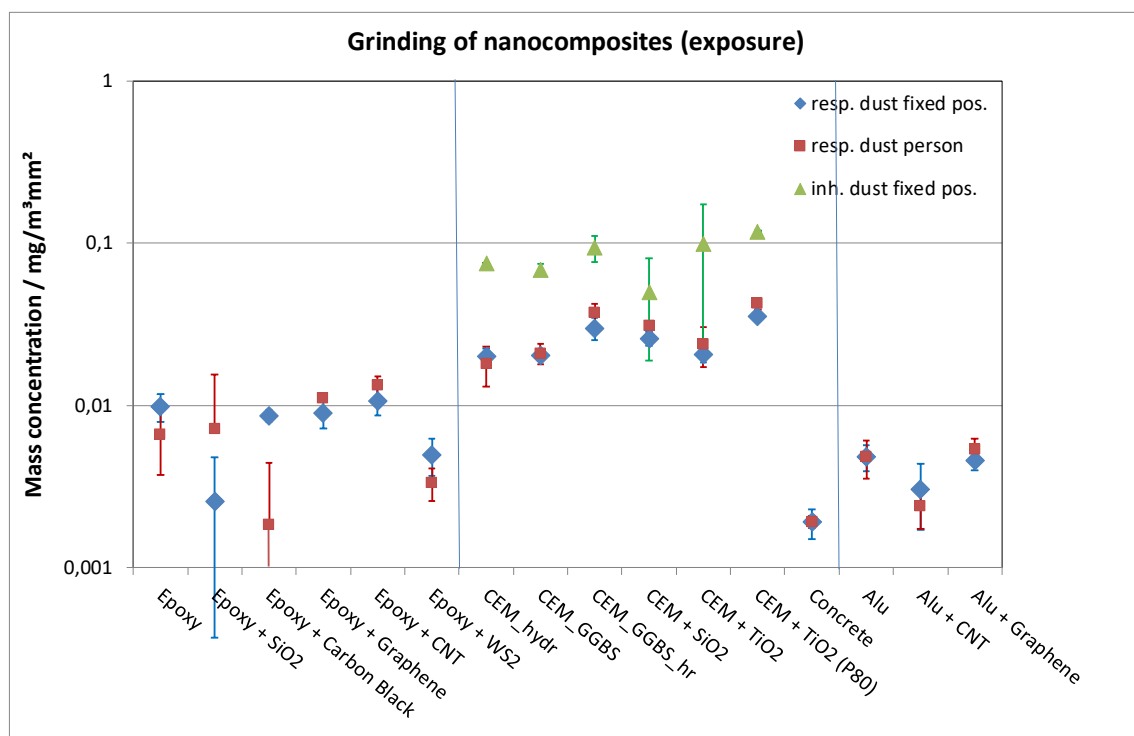


Fig.: Mass concentrations normalised to the area ground on the composite pieces

P-9 DOES EXPOSURE TO INFLAMMATORY PARTICLES MODIFY THE PATTERN OF ANIONS IN EXHALED BREATH CONDENSATE?

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Exposure to environmental and occupational particulate matter (PM) is recognized to induce health effects on the cardio-pulmonary system. In addition, associations between exposure to PM and metabolic syndromes like diabetes mellitus or obesity are now emerging in the literature. Collection of exhaled breath condensate (EBC) is an appealing non-invasive technique to sample pulmonary fluids.

This hypothesis generating study aims to (1) validate a ion chromatography method allowing the robust determination of different metabolism-related molecules (lactate, formate, acetate, propionate, butyrate, pyruvate, nitrite, nitrate) in EBC; (2) apply this method to EBC samples collected from workers exposed to quartz (a known inflammatory particle) and to soapstone (an a priori “inert” particle) as well as to controls.

A multi-compound standard solution was used to determine the linearity range, detection limit, repeatability and bias from spiked EBC. The biological samples were injected without further treatment into a ion chromatograph with conductivity detector. RTube® were used for field collection of EBC from 11 controls and workers exposed to soapstone (55 volunteers) or quartz dust (12 volunteers). The analytical method used proved to be adequate for quantifying eight anions in EBC samples. Its sub-micromolar detection limits and repeatability, combined with a very simple sample preparation, allowed an easy and fast quantification of different glycolysis or nitrosative stress metabolites. Using multivariate discriminant analysis to maximize differences between groups, we observed a different pattern of anions with a higher formate/acetate ratio in the EBC samples for quartz exposed workers compared to the soapstone group (Figure 1).

We hypothesize that a modification of the metabolic signature is induced by exposure to inflammatory particles like quartz and might be observed in the EBC via a change in the formate/acetate ratio.

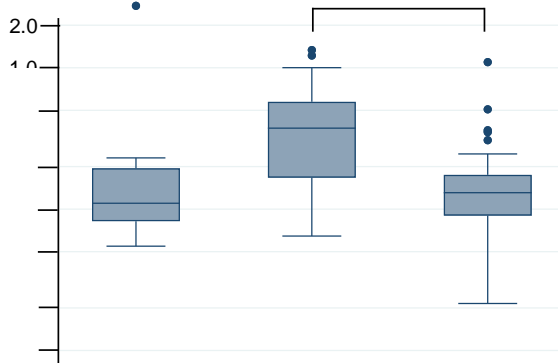


Figure 1: Box plot of the ratio formate/acetate for the three groups

An increase of formate could result from the reprogramming of the tricarboxylic cycle when cells are under oxidative stress. This hypothesis is plausible as some papers already reported modifications in the metabolic response of cells or animals exposed to different nanoparticles.

P-10 EXPOSURE TO UNINTENTIONAL NANOPARTICLES DURING THERMAL SPRAYING

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Protective coatings for metal structures used against corrosion and wear are frequently applied by thermal spraying. This study aims to assess exposure to unintentionally-released nanoparticles during thermal spraying at industrial scale under real-world conditions, by means of Atmospheric Plasma Spraying (APS) and High Velocity Oxy-Fuel spraying (HVOF). Particle monitoring was performed simultaneously: (a) inside the plasma booth in terms of particle number and mass concentration, mean diameter, TEM sampling for offline analysis; and (b) in the worker area outside of the plasma booth, in terms of particle size and mass distribution. In both processes the worker operated inside the plasma booth. Additionally, the APS was operating with doors closed while the HVOF had the door open. In both exposure scenarios the concentrations in terms of particle number (N) and mass were high inside the plasma booths and of the same order of magnitude in terms of N and mean particle diameter ($N = 10^6 \text{ cm}^{-3}$; 30-35 nm). HVOF had a major impact on worker area concentrations (Figure 1), as nanoparticles were transported through the open door to the rest of the facility, despite the extraction system. On the contrary, in the case of APS no significant fugitive emissions were observed due to the door sealing and the extraction system.

The main conclusions extracted from this study were:

- High concentrations in terms of N and PM_{10} were registered inside the plasma booths while the workers were operating.
- In the APS scenario nanoparticle emissions had low impact on worker exposure.
- In the HVOF scenario the doors were frequently open, resulting in impacts on nanoparticle exposures in adjacent areas.

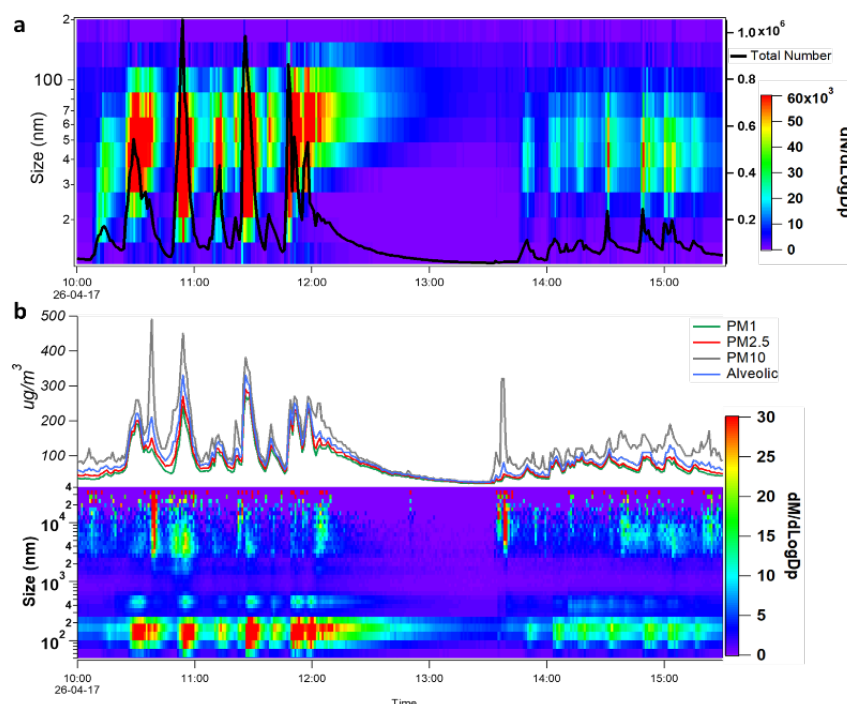


Figure. HVOF Time-series of total particle number concentration and size distribution (a); PM mass and mass distribution (b) in the worker area.

P-11 ANALYSIS OF TITANIUM DIOXIDE NANOPARTICLES IN FOOD BY SINGLE PARTICLE ICP-MS/MS

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Titanium dioxide (TiO₂) that is classified as food additive E171 within the European Union (EU) is added in different foodstuffs as a whitening and brightening agent. The food additive E171 mainly consists of micro-sized TiO₂ particles, but a certain nano-sized fraction (particles smaller than 100 nm) can still be present in the food. According to the EU Regulation 169/2011, engineered nanomaterials should be labelled as ingredients in food. This poses several analytical challenges in relation to sample preparation, the limitations of existing analytical techniques and the lack of validated studies and reference materials.

One of the most commonly used technique for the detection and characterization of metal-based nanoparticles (NPs) in food is inductively coupled plasma-mass spectrometry (ICP-MS) in single particle mode (spICP-MS). Measuring titanium at the most abundant isotope (m/z 48) is not possible due to isobaric interference from calcium at m/z 48 that cannot be removed with single quadrupole instruments. For calcium-rich samples, MS/MS technology in combination with reaction gases is therefore required.

In that regard, we investigated food samples containing TiO₂ as the white color additive E171. We applied spICP-MS technique for the analyses of TiO₂NPs in chewing gum, chocolate candy, cake frosting and in milk spiked with TiO₂NPs. For this purpose, spICP-MS analyses were performed on Agilent 8900 ICP-QQQ-MS in MS/MS mass-shift mode (m/z 48 \rightarrow m/z 64) with the use of O₂ and H₂ reaction gases in order to overcome isobaric interference of calcium.

Repeatable determination of number-based particle size distributions was achieved by spICP-MS with a minimum detectable particle size of ~30 nm. All the investigated food samples contained NPs (particles smaller than 100 nm) with the median diameters in the range of 130 to 160 nm. Similar size distributions for TiO₂NPs in different foodstuffs studied were also observed by asymmetric flow field flow fractionation (AF4) coupled to ICP-MS.

We showed that the use of O₂ and H₂ as reaction gases operated in MS/MS mode is able to efficiently remove the interferences of titanium on m/z 48 in matrices containing high concentration of calcium, such as milk. Together with short dwell time, this makes spICP-MS a promising technique for routine analysis of TiO₂ NPs down to ~30 nm in different food samples.

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